C T F A

THE COSMETIC, TOILETRY, AND FRAGRANCE ASSOCIATION

January 10, 2003

Dr. Michael Shelby
Director CERHR
NIEHS
79 T.W. Alexander Drive
Building 4401, Room 103
P.O. Box 12233, MD EC-32
Research Triangle Park, NC 27709

JAN 13 2003

E. EDWARD KAVANAUGH
PRESIDENT

RE: Request for Comments on the Draft Center for the Evaluation of Risks to Human Reproduction (CERHR) Expert Panel Report on Propylene Glycol

Dear Dr. Shelby,

The Cosmetic, Toiletry, and Fragrance Association (CTFA)[1] appreciates the opportunity to provide comments on the draft CERHR Expert Panel Report on Propylene Glycol. Propylene Glycol is used in personal care products, and thus, the consideration of Propylene Glycol by a CERHR Expert Panel is of interest to CTFA members.

The Cosmetics Industry supports an independent Expert Panel, the Cosmetic Ingredient Review (CIR), that evaluates the safety of ingredients as they are used in cosmetic/personal care products. The CIR Expert Panel is comprised of seven voting physicians and scientists and includes representatives of the U.S. Food and Drug Administration, the Consumer Federation of America, and the Cosmetics Industry as non-voting liaison members. A recent Annual Report is enclosed.

CIR has evaluated the safety of Propylene Glycol. The CIR Expert Panel concluded that "Propylene Glycol and Polypropylene Glycols are safe for use in cosmetic products at concentrations up to 50.0%" (JACT 13(6): 437-491. 1994). A copy of the CIR report on Propylene Glycol is enclosed. The CIR report provides information about the product categories and concentration ranges of Propylene Glycol used in cosmetic/personal care products that is lacking in the draft CERHR Expert Panel Report on Propylene Glycol.

We trust you will find the information in the CIR report helpful to the CERHR review of Propylene Glycol, and encourage you to contact us should you need additional information.

Sincerely,

Jerry McEwen

Gerald N. McEwen, Jr., Ph.D., J.D.
Vice President - Science

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[1]CTFA is the U.S. national trade association representing the personal care products industry. CTFA is comprised of over 300 active members that produce the vast majority of the cosmetics distributed in the U.S. and that also produce many over-the-counter drugs designed for dermal application. The association also has over 300 associate members that provide raw ingredients and other supplies and services to the industry. Many of CTFA's members are international companies that do business in many foreign countries as well.

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Final Report on the Safety Assessment of Propylene Glycol and Polypropylene Glycols[1]

Abstract: Propylene Glycol is an aliphatic alcohol manufactured as a reaction product of propylene oxide and water. Polypropylene Glycol is a polymer formed by adding propylene oxide to dipropylene glycol. Propylene Glycol is reportedly used as a skin-conditioning agent–humectant, solvent, viscosity-decreasing agent, and humectant in thousands of cosmetic formulations. Polypropylene Glycols of various polymer lengths are reportedly used as miscellaneous skin-conditioning agents in far fewer formulations. Acute, subchronic, and short-term animal studies suggested little toxicity beyond slight growth and body weight decreases. Little ocular or skin irritation was observed in animal studies, and no sensitization was seen. Small increases in fetal malformations were seen in mice injected subcutaneously with Propylene Glycol, but a continuous breeding reproduction study in mice showed no reproductive toxicity following oral administration. A wide range of mutagenesis assays were negative, and studies in mice and rats showed no evidence of carcinogenesis. Clinical data showed skin irritation and sensitization reactions in Propylene Glycol in normal subjects at concentrations as low as 10% under occlusive conditions and dermatitis patients at concentrations as low as 2%. A careful evaluation of skin irritation and sensitization data as a function of disease state of the individual, occlusion, and concentration was done. On the basis of that analysis, it is concluded that Propylene Glycol and Polypropylene Glycol are safe for use in cosmetic products at concentrations up to 50%. Key Words: Propylene Glycol—Polypropylene Glycol.

CHEMISTRY

Definition and Structure

Propylene Glycol (PG) is an aliphatic alcohol that conforms to the following formula (Estrin et al., 1982*a*):

$$CH_3CHCH_2OH$$
$$\quad\quad|$$
$$\quad\quad OH$$

PG (CAS No. 57-55-6) is also known as 1,2-Propanediol, 1,2-Dihydroxypropane, Methylethylene Glycol, Methyl Glycol, Monopropylene Glycol, Pro-

[1] Reviewed by the Cosmetic Ingredient Review Expert Panel.
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pane-1,2-Glycol, α-Propyleneglycol, 1,2-Propylene Glycol, and numerous trade names (Estrin et al., 1982*a*; Sweet, 1987).

Polypropylene Glycol (PPG) is a polymer of PG and water that generally conforms to the following formula (Estrin et al., 1982*a*; Food Chemicals Codex, 1981):

$$HO(C_3H_6O)_nC_3H_6OH$$

n = number of oxypropylene groups

Polypropylene Glycol (CAS No. 25322-69-4) is also known as Polyoxypropylene, PPG, and numerous trade names (Estrin et al., 1982*a*).

Properties

The chemical and physical characteristics of PG and PPG are listed in Tables 1 and 2, respectively.

Analytical Methods

PG has been analyzed via gas chromatography (Gebhardt, 1968; Demey et al., 1988) and ultraviolet (UV) spectral analysis (Dow Chemical Co., 1992; Eastman

TABLE 1. *Chemical and physical properties of Propylene Glycol*

		Ref.
Physical form	Colorless, hygroscopic liquid	Estrin et al. (1982*b*)
Odor	Practically odorless	Sax (1979)
Formula	$C_3H_8O_2$	Estrin et al. (1982*a*)
Molecular weight	76.11	Sweet (1987)
		Sax (1979)
Assay	97.5% minimum (cosmetic use)	Estrin et al. (1982*b*)
	99.5% minimum (industrial use)	Dow (1975, 1978)
Vapor density	2.62 (air = 1)	Sax (1979)
Vapor pressure	0.08 mm Hg at 20°C	Sax (1979)
Refractive index	1.4324 at 20°C	Sax (1979)
Density	1.0361 at 25°/4°C	Weast (1982)
	1.0362 at 25°/25°C	Sax (1979)
Specific gravity	1.035–1.037 at 25°/25°C	Estrin et al. (1982*b*)
Boiling point	189°C	Weast (1982)
	188.2°C	Sax (1979)
Freezing point	−59°C	Sax (1979)
Flash point	210°F (open cup)	Sax (1979)
Autoignition temperature	700°F	Sax (1979)
Solubility	Soluble in water, ethanol, benzene	Weast (1982)
Distillation range	184–189°C	Estrin et al. (1982*b*)
Acidity	0.002 mEq/ml maximum	Estrin et al. (1982*b*)
Sulfated ash	0.07% maximum	Estrin et al. (1982*b*)
Water	0.2% maximum	Estrin et al. (1982*b*)
Arsenic (as As)	3 ppm maximum	Estrin et al. (1982*b*)
Explosion hazard	Moderate when exposed to flame	Sax (1979)
Lower explosion limit	2.6%	Sax (1979)
Upper explosion limit	12.6%	
Fire hazard	Low	Sax (1979)

TABLE 2. *Chemical and physical properties of Polypropylene Glycol*

		Ref.
Physical form	Clear, colorless, viscous liquid	Food Chemicals Codex (1981)
Formula	$HO(C_3H_6O)_nH$	Sax (1979)
Molecular weight	400–2,000	Sax (1979)
Density	1.002–1.007	Sax (1979)
Flash point	390 to >440°F	Sax (1979)
Solubility	Soluble in water, aliphatic ketones, and alcohols; insoluble in ether and aliphatic hydrocarbons	Food Chemicals Codex (1981)
Viscosity	85–97 centistokes at MW = 1,000	Food Chemicals Codex (1981)
	150–175 centistokes at MW = 2,000	Food Chemicals Codex (1981)
Arsenic (as As)	3 ppm maximum	Food Chemicals Codex (1981)
Heavy Metals (as Pb)	5 ppm maximum	Food Chemicals Codex (1981)
pH	Between 6 and 9	Food Chemicals Codex (1981)
Propylene oxide	0.02% maximum	Food Chemicals Codex (1981)
Residue on ignition	0.01% maximum	Food Chemicals Codex (1981)
Fire hazard	Slight when exposed to heat or flame, may react with oxidizing materials	Sax (1979)

Chemical Co., 1992; Istituto Ganassini spa di ricerche biochimiche, 1992). Each spectral analysis indicates no absorbance in the UVA and UVB bands.

PPG 1200 has been detected at sub-ppm levels in aqueous and organic media by a method that combines the classic Zeisel alkoxy reaction with high-efficiency gas-liquid chromatography (Ramstad et al., 1978).

Methods of Manufacture

All PG that is of commercial significance is manufactured as a reaction product of propylene oxide and water; no additives or solvents are used. Additionally, the chlorohydrin process is used to manufacture the reactant, propylene oxide, which is recovered as a pure product before conversion to glycol (Dow U.S.A., 1993).

PG is also manufactured by treating propylene with chlorinated water to form the chlorohydrin; the chlorohydrin is converted to the glycol by treatment with sodium carbonate solution. Another method of preparation involves heating glycerol with sodium hydroxide (Rothschild, 1988).

PPGs are manufactured by the addition of propylene oxide to dipropylene glycol (Shaffer et al., 1951).

Impurities

In cosmetic products, the purity of PG is specified as a minimum of 97.5%. In industry, the purity of PG is specified as a minimum of 99.5%. Impurities found in PG may include sulfated ash, arsenic, propylene oxide, and heavy metals such as lead (Estrin, et al. 1982*b;* Food Chemicals Codex, 1981). The specific maximum levels of impurities that may be present in both food- and cosmetic-grade PG and PPG are listed in Tables 1 and 2.

The Dow Chemical Co. (Dow U.S.A.) recommends that USP-grade PG be used for cosmetics. USP-grade PG has a typical assay of 99.9% and a specification of

TABLE 3. *Product formulation data for Propylene Glycol (FDA, 1984)*

Product category	No. of product formulations within each concentration range (%)								
	>50	25–50	10–25	5–10	1–5	0.1–1	0–0.1	Unknown	Total
Baby shampoos				1	2	1		3	7
Baby lotions/oils/powders/creams				1	5			2	8
Other baby products					2	1			3
Bath oils/tablets/salts		2	4		2	5	13	3	29
Bubble baths			1	6	15		64	37	123
Other bath preparations		2	1	1	14	5	20	5	48
Eyebrow pencil					1				1
Eyeliner			2	7	42		4		55
Eye shadow			3	71	18	44		39	175
Eye lotion				1	2		1		4
Eye makeup remover		2	2	1	8	4			17
Mascara			4	6	30	4	2	14	60
Other eye makeup preparations				13	14		7	9	43
Colognes/toilet waters		5		8	27	16	40		96
Perfumes		1		3	4	7	13		28
Powders dusting/talcum excl. aftershave talc					1	4	4	1	10
Sachets		4	6		14	2	2		28
Other fragrance preparations	1	4		9	19	4	6		43
Hair conditioners				7	17	10	10	14	58
Hair sprays (aerosol fixatives)					1	6	2	1	10
Hair straighteners				10	12				22
Permanent waves		3		2		25	2	11	43
Rinses (noncoloring)				1	3	1	5	3	13
Shampoos (noncoloring)			4	10	60	52	31	54	211
Tonics/dressings/other hair grooming aids			2	5	13	2	6	3	31

Wave sets		1	1	5	5	6				18
Other hair preparations	1	1	1	5	7			1		16
Hair dyes/colors (requiring caution statement)		60	140	82	1			5		288
Hair rinses (coloring)			4	18	7					29
Hair shampoos (coloring)			1			2				3
Hair lighteners with color				1						1
Hair bleaches			2	4						6
Other hair coloring preparations	1			3				1		5
Blushers (all types)		3	31	14	7	12		17		85
Face powders	1			16	1	12				29
Foundations		45	106	44	1	10		56		262
Leg and body paints				3						3
Lipstick			1	3	244	389		544		1181
Makeup bases		12	200	61	2	7		52		334
Rouges		2	7	4	2	7		8		30
Makeup fixatives					1			3		4
Other makeup preparations	1	3	8	17	4	27		41		131
Cuticle softeners			6	3	1	2				12
Nail creams/lotions				6	1					7
Nail polish and enamel removers					1	1				2
Other manicuring preparations		2		3						6
Dentifrices (aerosol/liquid/paste/powder)		1		1						2
Mouthwashes/breath fresheners				3						3
Other oral hygiene products						1				1
Bath soaps/detergents				5	6	4	24			39
Deodorants (underarm)	19	4	9	28	43	10	2	9		124
Douches		1	4		1		1			7
Feminine hygiene products				1		1				2
Other personal cleanliness products		3		9	24	16	1			53
Aftershave lotions		1		5	49	28	8	6		97

(Continued)

TABLE 3. *Continued*

Product category	No. of product formulations within each concentration range (%)								
	>50	25–50	10–25	5–10	1–5	0.1–1	0–0.1	Unknown	Total
Beard softeners			2					1	3
Preshave lotions					1	1	2	4	8
Shaving cream (aerosol/brushless/lather)			2	1	17	8	1	5	34
Other shaving preparations			1	2	3	5		2	13
Skin cleansing products (cold creams/ lotions/liquids/pads)		2	15	67	128	12	23	29	276
Depilatories			2	2			2		6
Face/hand/body (excl. shaving preparations)		1	14	71	197	48	31	55	417
Foot powders/sprays					1				1
Hormone products			1	1	2		1		5
Moisturizing products		2	5	99	170	18	40	24	358
Night preparations		1	6	30	50	7	6	5	105
Paste masks (mud packs)			5	14	45	7	2	10	83
Skin lighteners			2	6	9			2	19
Skin fresheners			1	10	56	17	15	37	136
Wrinkle-smoothing products (removers)			1	2	6	2	2	1	14
Other skin care preparations		1	4	25	55	18	14	32	149
Suntan gels/creams/liquids			2	14	30	3	12	15	76
Indoor tanning preparations				1	9			2	12
Other suntan preparations			1	4	5		1	4	15
Ingredient total	21	43	236	1,068	1,529	683	896	1,200	5,676

99.5% minimum purity. Dow U.S.A. recognizes that the United States Pharmacopeia now allows up to 5 ppm propylene oxide in PG and is of the opinion that typical levels contained in products today are less than detectable amounts (Dow U.S.A., 1993).

COSMETIC USE

PG is used as a skin-conditioning agent–humectant, solvent, viscosity-decreasing agent, and humectant in cosmetic formulations (Nikitakis, 1988). Data on the uses of PG are summarized in Table 3. Data submitted to the Food and Drug Administration (FDA) in 1984 indicated that PG was used in a total of 5,676 cosmetic formulations. Use concentrations of PG in cosmetic formulations are listed in Table 3 as <0.1 to >50% (FDA, 1984).

PPGs are used as miscellaneous skin-conditioning agents in cosmetic formulations (Nikitakis, 1988) as listed in Tables 4–6. Data on the uses of three of the PPGs, PPG 9, PPG 26, and PPG 425, indicate that PPG-9 has been used in a total of 6 cosmetic formulations at concentrations of 1–5 or 0.1–1% and PPG 26 in a total of 10 cosmetic formulations. The largest use of PPG 26 was in blusher products at a concentration of 1–5%. Table 6 indicates that PPG 425 was used in one cosmetic formulation, a hair bleach, at a concentration of between 1 and 5% (FDA, 1984).

Voluntary filing of product formulation data with the FDA by cosmetic manufacturers and formulators must conform to the format of concentration ranges and product categories as described in Title 21, Part 720.4 of the Code of Federal Regulations (21 CFR 720.4). Since certain cosmetic ingredients are supplied to the formulator at <100% concentration, the concentration reported by the formulator may not necessarily reflect the actual concentration found in the finished cosmetic product; the actual concentration in such an instance would be a fraction of that reported to the FDA. The fact that data are submitted only within the framework of a "concentration range" provides the opportunity for overestimation of the actual concentration of an ingredient in a particular product. An entry at the lowest end of a concentration range is considered the same as one entered at the highest end of that range, thus introducing the possibility of a 2- to 10-fold error in the assumed ingredient concentration.

Concentration of use data are no longer reported to FDA by the cosmetics industry (Federal Register, 1992). However, data on PG supplied to the Cosmetic, Toiletry, and Fragrance Association (CTFA) indicate that concentrations of use

TABLE 4. *Product formulation data for PPG 9 (FDA, 1984)*

	No. of product formulations within each concentration range (%)							
Product category	25–50	10–25	5–10	1–5	0.1–1	0–0.1	Unknown	Total
Other bath preparations				2				2
Shampoos (noncoloring)				2	2			4
Ingredient total				4	2			6

TABLE 5. *Product formulation data for PPG 26 (FDA, 1984)*

Product category	No. of product formulations within each concentration range (%)							
	25–50	10–25	5–10	1–5	0.1–1	0–0.1	Unknown	Total
Bath oils/tablets/salts				1				1
Blushers (all types)				3				3
Deodorants (underarm)				2	2			4
Moisturizing products				1				1
Other skin care preparations				1				1
Ingredient total				8	2			10

range between 3 and 5% in current products manufactured by one company. The intention of using PG in cosmetics at concentrations as great as 80% was also stated (CTFA, 1992). Current frequency of use data indicate that PG is used in a total of 4,056 cosmetic products. PPG 9 is used in 13 products, and PPG 12 and PPG 26 are used in 2 and 5 products, respectively (FDA, 1993).

Products containing PG or PPG may contact the skin, hair, and eyes. Ingestion of small quantities of either compound is possible. Contact with either compound may be as little as once to as often as daily for several hours.

International Use

PG, PPG 9, and PPG 26 are included in the list of ingredients that have been approved for use in cosmetic formulations marketed in Japan (Nikko Chemicals, Co., Ltd., 1991–92). PG and PPGs are not included in the list of ingredients that are prohibited from use in cosmetic products marketed in the European Economic Community (EEC Cosmetics Directive, 1990).

NONCOSMETIC USE

PG is generally recognized as safe (GRAS) when used in accordance with good manufacturing practices (21 CFR 582.1666). A monograph prepared for the GRAS evaluation is available (Informatics, 1973). PG is also an accepted optional ingredient in standardized food substances such as vanilla extract (21 CFR 169.175).

In an assessment of food additives, the World Health Organization (WHO) recommended a maximum daily oral intake of PG of 25 mg/kg body wt/day (WHO, 1974).

PG is an ingredient in Over-the Counter (OTC) drug products (FDA, 1991).

TABLE 6. *Product formulation data for PPG 425 (FDA, 1984)*

Product category	No. of product formulations within each concentration range (%)							
	25–50	10–25	5–10	1–5	0.1–1	0–0.1	Unknown	Total
Hair bleaches				1				1
Ingredient total				1				1

FDA issued a proposed rule on the use of PG as a pediculicide, indicating that OTC drug products containing PG for this purpose are not generally recognized as safe and effective or are misbranded. An advance notice of proposed rulemaking containing a similar statement regarding use of PG as a hair grower was also issued by FDA. OTC drug products containing PG for use as a demulcent are generally recognized as safe and effective and are not misbranded; this statement constitutes final agency action (FDA, 1991). PG has been classified as both an excellent solvent and an effective preservative; it has replaced glycerin as the vehicle in many formulations of topical therapeutic agents (AMA Division of Drugs, 1983).

Other documented noncosmetic uses of PG include use of the compound in the production of varnishes and resins and as an antifreeze (Angelini and Meneghini, 1981). PG has been used as a source of energy in animal diets (Emmanuel, 1976), as a cryoprotective agent in renal preservation (Pegg et al., 1987; Halasez and Collins, 1984), and as a therapy for ketosis in cattle (Ruddick, 1972).

An investigation of PG as a potential environmental contaminant is available (National Technical Information Service, 1979).

PPG is an indirect food additive that has been approved for use as a component of resinous and polymeric coatings (21 CFR 175.300).

GENERAL BIOLOGY

Absorption, Metabolism, and Excretion

Propylene Glycol

In mammals, the major route of PG metabolism is to lactaldehyde and then lactate via hepatic alcohol and aldehyde dehydrogenases; in an alternative pathway, lactaldehyde is metabolized to methylglyoxal before lactate is formed (Christopher et al., 1990*a*).

A study on the metabolism of PG was conducted by Morshed et al. (1988). Five groups of six rats each were given oral doses of 4.83, 9.66, 19.32, 38.64, and 77.28 mmol of aqueous PG/kg body wt. Blood was collected from these animals at times ranging from 5 min to 24 h postdosing. Appearance of PG in the blood of the treated rats was in a dose-related manner. The maximum concentration of PG in the blood, 28 mmol/L, was found 2 h after dosing in the rats given the largest dose.

In the second part of this experiment, the metabolism of PG was further investigated. The metabolism of PG in animals is initially catalyzed by several dehydrogenases. To test inhibition of PG metabolism, the authors pretreated five groups of six Wistar rats each with oral doses of 0.0, 0.025, 0.05, 0.20, and 1.0 mmol pyrazole, a known alcohol dehydrogenase inhibitor. The rats were then given oral doses of 4.83, 9.66, 19.32, 38.64, or 77.28 mmol/kg PG. Blood samples were collected in a manner similar to that in the preceding experiment. In the pretreated rats, the greatest blood concentrations of PG were found in the blood 30 min postdosing, regardless of the amount of compound administered. The peak concentration of PG in the blood of these animals was significantly reduced compared with rats dosed with PG that were not pretreated with pyrazole.

In a separate experiment, the same authors tested the urinary clearance of PG from rats pretreated with pyrazole. Three groups of rats, 36 per group, were given oral doses of 0.0, 0.20, or 1.0 mmol/kg pyrazole. Each group was subdivided into three subgroups (a total of nine groups) and given 19.32, 38.64, or 77.28 mmol/kg PG orally. The excretion of PG in the urine increased according to the amount of PG administered, both with and without pyrazole. The amount of PG excreted was significantly greater in the rats fed doses of pyrazole then in the control rats.

The metabolism of PG in the rabbit was investigated by Yu and Sawchuk (1987). Three groups of New Zealand White male rabbits, three animals per group, were given intravenous doses of 0.5, 1.0, or 2.0 g/kg PG in normal saline solution. Blood and urine were collected between 0 and 12 h postdosing. The plasma concentration of PG was measured, and the half-lives of the compound in the blood were calculated. In rabbits given the smallest dose, the half-life was 64.0 min. For the 1.0- and 2.0-g/kg doses, the half-lives were 76.2 and 75.4 min, respectively. As the plasma concentration of PG diminished, the metabolism clearance of PG increased proportionally. It was determined that there was a nonlinear relationship between the two routes of clearance of PG.

After oral administration of PG (38.66 mmol/kg, single dose) to four adult New Zealand rabbits (weights 2–2.5 kg), the concentration of L-lactate in whole blood increased significantly ($p < 0.01$, $n = 4$) to 2.55 ± 0.62 mmol/L when compared with a fasting concentration of 1.04 ± 0.22 mmol/L. The concentration of D-lactate was also significantly elevated ($p < 0.001$). D- and L-Lactate are PG metabolites. Blood concentrations of pyruvate remained mostly unaffected both before and after PG administration (Morshed et al., 1991).

Speth et al. (1987) investigated the effects of PG administration using human subjects. Six patients with confirmed malignancies were given intravenous doses of a potentially cytostatic agent, mitoquidone, dissolved in PG. Subjects received doses of 5.1–21.0 g PG/day in a 4-h intravenous infusion. The concentration of PG in the plasma and the half-life of PG in the body were measured. The average half-life was 2.3 ± 0.7 h, and the elimination of PG from the body was in a dose-related manner. The authors also measured lactic acid concentration, venous pH, plasma osmolality, hemoglobin, and haptoglobin levels in each test subject. No significant alterations in any of these parameters were noted.

Polypropylene Glycols

PPG 425 and PPG 1025 are readily absorbed from the gastrointestinal (GI) tract. Following administration of a single oral dose of PPG 425 (0.5–1.0 g/kg) to rabbits, 17–39% was recovered from the urine. Under similar conditions of exposure, ~40% of administered PPG 1025 was excreted in the urine of rabbits. After rabbits received a single oral dose of PPG 2025 (2 g/kg), 40–70% was excreted in the feces and a negligible amount was excreted in the urine. For all of the groups tested, neither the weight range, strain, nor number of animals involved was stated (Shaffer et al., 1951). Dermal absorption of PPG 425 has been demonstrated in rabbits (Shaffer et al., 1951) (see Acute Dermal Toxicity).

Cellular Effects

The effects of PG on mouse urinary bladder epithelial cell growth were investigated by Farsund (1981). A group of hairless male Oslo strain mice (number not specified) was given subcutaneous injections of 0.2 ml PG 3 days a week for 3 months. There were no significant changes in the fraction of cells with S-phase DNA content among the diploid cells. Cells harvested after 1 month of treatment with PG had a 50% reduction in the DNA production in tetraploid cells. DNA production was reduced to 20% after 2 months of treatment and was 0% at 3 months. The number of tetraploid cells was nearly 0 at the end of the 3-month treatment period. PG caused a disturbance in the proliferative response of rat urinary bladder epithelial cells.

Mochida and Gomyoda (1987) investigated the effects of doses of PG on cultured human KB cells. According to methods described by Mochida et al. (1983), the 50% inhibitory dose of PG was 0.31 *M* in cells incubated with the compound for 72 h.

Immune System Effects

Denning and Webster (1987), studied the in vitro effects of PG on human natural killer cell and neutrophil cell populations. In the first half of the experiment, a natural killer cytotoxicity assay was performed. Peripheral mononuclear cells were isolated from the blood of one human volunteer and used as the effector cells in the assay. Cultured K562 erythroleukemia cells were used as the target cells. Three concentrations of effector cells were incubated in separate wells of microtiter plates with PG in phosphate-buffered saline (PBS) diluted to final concentrations of 0.01, 0.1, and 1%. PBS alone was used as a control. One hundred microliters of the target cells was added to each well. Cytotoxicity was measured by observing the percentage ^{51}Cr release. The cytotoxicity of human natural killer cells was decreased significantly when cells were incubated with 1% PG ($p < 0.002$). Concentrations of 0.01 and 0.1% PG did not significantly decrease the natural killer cell activity.

In the second part of the experiment, the same authors tested the effects of PG on isolated human neutrophils. Blood was collected from a human volunteer, and mononuclear cells were isolated. The isolated cells were incubated in a solution containing Hanks' Balanced Salt Solution, heparin, and 10 µl luminol (5-amino-2,3-dihydro-1,4-phthalazimedione). The incubated cells were taken in aliquots of 1 ml and placed in specially designed cuvettes for 30 min. The cells were then incubated with PG diluted to final concentrations of 0.0, 0.1, 0.5, or 1.0%. Latex particles coated with IgG were added to each tube, and the chemiluminescence was measured. The neutrophil chemiluminescence was significantly decreased in the cells incubated with 0.5 and 1% PG.

Other Biological Effects

Floersheim (1985) investigated the effects of PG on the metabolism of ethanol in mice. Four groups of four female albino MAG mice were injected with 0.0, 0.05

ml/10 g, 0.025 ml/10 g, or 0.01 ml/10 g PG 30 min prior to the intraperitoneal administration of 4,500 mg/kg body wt ethanol. The survival rate for the mice given 0.01 ml PG was 19%. The survival rates in the mice fed the two larger doses were 75% for the mice given 0.05 ml PG and 87% for the mice given 0.025 ml PG. The survival rates in the mice given the two larger doses were significantly greater ($p < 0.001$) than in the mice given the lowest dose.

The effects of PG on the metabolism of enflurane anesthesia were investigated by Fish et al. (1988). Thirty-two male Fischer strain rats were divided into four groups. Saline was administered to 10 control rats, 10 rats received 0.4 mg/ml etomidate, 6 received 7% (vol/vol) PG intravenously, and 6 received PG plus etomidate. All rats were then exposed to 2% enflurane vapor in an anesthesia chamber for a period of 1 h. Serum samples were collected at 0, 2, 4, and 24 h after exposure to the vapor. The metabolism of the anesthesia was measured by analysis of rat serum F^-. There was a significant decrease in the rate of metabolism of enflurane in rats pretreated with PG as compared with saline controls ($p < 0.05$).

In a second experiment, the same authors tested the effects of PG administration on adrenal steroidogenesis in the rat. Four groups of male Fischer strain rats, five per group, were given intravenous injections of saline (controls), 0.4 mg/ml etomidate, 7% (vol/vol) PG, or etomidate plus PG. Thirty minutes later, 0.2 μg adrenocorticotropic hormone (ACTH) was administered to all groups via intravenous injection, causing aldosterone to be released into the serum. Serum samples were drawn at 30, 60, and 90 min post-ACTH administration. Administration of PG did not cause any significant changes in adrenal steroidogenesis in the rat in this experiment.

The effects of intraperitoneal doses of PG on the metabolism of Hexobarbital and Zoxazolamine in the rat were studied by Dean and Stock (1974). Two groups of rats, three per group, were given doses of 4 ml/kg PG twice a day for 3 consecutive days. Two groups of three rats each served as untreated controls. Each group was then given either 125 mg/kg Hexobarbital (to test effect on sleeping time) or 120 mg/kg Zoxazolamine (to test effect on paralysis time). The rats pretreated with PG and given Hexobarbital had significantly greater sleeping times than the control rats ($p < 0.01$). The treated rats given Zoxazolamine had significantly longer paralysis times than the control rats ($p < 0.01$).

The effects of PG on antipyrine metabolism in humans were investigated by Nelson et al. (1987). Ten healthy volunteers were given 5-ml doses of PG orally 10 min prior to administration of 1.2 g antipyrine. Additional doses of PG were administered every 4 h for 44 h (total PG dose = 55 ml). Plasma samples were taken from the venous cannula at 0, 5, 10, 15, 30, and 45 min and 1, 2, 4, 6, 8, 10, 12, 24, and 32 h after PG administration. Plasma antipyrine concentrations were measured by high-performance liquid chromatography. The authors found no significant effects on antipyrine clearance in humans due to PG administration at the doses investigated in this experiment.

Male Sprague–Dawley rats (number not stated) were given 4 ml/kg/day PG via gastric intubation for 30 days in a short-term study by Hoenig and Werner (1980). A second group of animals was given distilled water and served as the control.

Body weights were measured throughout the experiment. On day 31, blood samples were taken from the carotid artery, and the livers of the animals were removed for lipid analysis. There were no significant differences in body weights or liver weights in the PG-treated animals as compared with controls. There was a significant increase in total cholesterol in the livers of the PG-treated animals ($p = 0.02$).

The effect of PG on liver structure was evaluated using 15 male Wistar rats (5 weeks old). PG [15% (vol/vol) in drinking water] was administered to the animals daily by gastric intubation, and animals were killed at time periods ranging from 1 week to 3 months after initiation of the study. Thin sections of the liver, fixed with glutaraldehyde, were examined using an electron microscope. PG administration resulted in remarkably enlarged mitochondria with well-developed cristae and a dense matrix in every hepatocyte. These changes were noted throughout the hepatic lobule, but were more distinct in hepatocytes of the peripheral zone of the hepatic lobule. Other ultrastructural changes in hepatocytes included proliferation of smooth-surfaced endoplasmic reticulum and an increase in the number of lysosomes and microbodies (Wakabayashi et al., 1991).

Enzyme effects of short-term administration of PG in rats were investigated by Amma et al. (1984). Two groups of six male and six female rats were given oral doses of 1 ml 28.4% PG/100 g body wt daily for 30 days. A control group of 12 rats was given daily doses of distilled water. On day 31, blood samples for enzyme studies were collected from the orbital sinus of each animal. Glucose-6-phosphate: NADP oxidoreductase, blood catalase activity, and whole-blood reduced glutathione (GSH) were measured. The growth rate of test animals was not significantly different from controls. In the treated animals, the whole-blood GSH was significantly decreased ($p \leq 0.001$), and erythrocyte catalase activity was significantly decreased in the males ($p \leq 0.01$).

Ahluwalia and Amma (1984) studied the effects of short-term administration of PG on rat erythrocyte morphology and function. One group of six male rats was given oral doses of 284 μl PG daily for 30 days. A second group of six rats serving as a control group was given oral doses of distilled water. On day 31, blood samples were drawn from the orbital sinus of each rat, and membrane lipids were extracted from erythrocytes. Short-term oral doses of PG caused a significant decrease in total lipids ($p < 0.001$), cholesterol ($p < 0.05$), and phospholipids ($p < 0.05$) in the rat erythrocytes. The decrease in lipids altered the erythrocyte morphology in the PG-treated animals.

Christopher et al. (1989) investigated the effects of oral PG administration on the development of Heinz body hemolytic anemia in cats. Two groups of cats were fed commercial diets in the conditioning phase of the experiment. Control values for Heinz bodies were obtained. Each cat served as its own control. Six of the cats were fed diets containing 12% (1.6 g/kg/day) PG for 5 weeks (low-dose study). Five adult cats were fed diets containing 41% (8 g/kg/day) PG for 22 days (high-dose study). Blood samples were collected and Heinz body formation was investigated. The percentage of Heinz bodies present during the control period was $2.98 \pm 0.90\%$ in the low-dose group. After 1 week on the 12% PG diet, the percentage of Heinz bodies was significantly increased to $11.33 \pm 3.36\%$ ($p <$

0.002). After 35 days, the percentage of Heinz bodies was 28%. In the high-dose group, the control value for percentage of Heinz bodies was 4.8 ± 1.81%. The percentage of Heinz bodies was 92.1 ± 3.6% on day 9. This value was significantly increased compared with control values ($p < 0.001$).

Singh et al. (1982) investigated various pharmacologic parameters of PG in a series of animal studies. Swiss mice (number not stated) in a locomotor activity experiment were given either intraperitoneal or oral doses of 10, 20, 50, or 100% PG. Locomotor activity was measured at 15, 30, 60, and 120 min postdosing. In animals given 100% PG intraperitoneally, there was a significant reduction in locomotor activity.

The effect of PG administration on forced locomotor activity was also studied in Wistar rats and Swiss mice (number not stated). Mice given peroral doses of 100% PG had a 60% reduction in activity compared with controls. Upon intraperitoneal administration of the compound, there was a significant decrease in forced locomotor activity in both rats and mice given 100% PG. With use of the same doses in an inclined plane test involving mice, a smooth wooden plate at an angle of 45° was used to determine ataxia. By both routes of administration, 20–100% PG caused ataxia in mice.

Further investigations included the effects of PG on rat and guinea pig smooth muscle samples. Pieces of the ileum from a guinea pig were placed in an organ bath and 0.5 ml of 10, 20, 50, or 100% PG was added for 3 min. Contractions were stimulated in the muscles by addition of acetylcholine chloride, histamine dihydrochloride, or barium chloride. At concentrations above 20%, a nonspecific, dose-dependent inhibition of contraction was noted. In the rat, uterine samples were immersed in organ baths and PG (same concentrations) were added. Contractions were induced in the smooth muscle samples by addition of 5-hydroxytryptamine. All PG concentrations produced a nonspecific dose-dependent inhibition of contractions. PG did not produce any paralytic effects, analgesic activity, or hypnotic effects in mice or rats at any of the concentrations tested.

Several other investigators also studied the effects of PG on muscle function. The potential for 40% (vol/vol) PG in distilled water to cause skeletal muscle damage after intramuscular injection was evaluated in vitro using isolated extensor digitorum longus muscles from rats. The muscles were injected with 15 μl of the solution and placed in a balanced salt solution bubbled with 95% O_2/5% CO_2. The myotoxicity of injected solutions was evaluated by noting the cumulative release of creatine kinase (intracellular cytosolic enzyme) into the incubation medium over a 2-h period. Creatine kinase levels and histological evaluation are the commonly used indexes of skeletal muscle damage, both clinically and experimentally. Cumulative creatine kinase release increased as a function of time throughout the 2-h incubation period (Brazeau and Fung, 1989).

The results of this study were then evaluated with in vivo studies on creatine kinase activity in five male New Zealand white rabbits (weights 2.0–4.2 kg). The animals received a 1-ml injection of each of the following: 40% (vol/vol) PG, 40% (vol/vol) polyethylene glycol 400, and normal saline. The solutions were injected into the mid-lumbar muscles of each animal in a three-way crossover random design; there was a minimum of 2 weeks between each phase of the experiment.

The injection sites randomly rotated such that no single muscle area received more than one injection, and blood samples were obtained from the central artery or marginal ear vein up to 72 h postinjection. The corrected area under the serum kinase activity versus time curve following propylene glycol injection was statistically larger ($p < 0.05$) than those after the injection of polyethylene glycol and normal saline (Brazeau and Fung, 1989).

Kaldor et al. (1971) studied the effects of PG and various PPGs on the contraction of rabbit muscles. Glycerol-extracted fibers of rabbit psoas muscle were cut into 50-mm bundles. Half of each bundle was incubated with 0.2 *M* KCl/0.05 *M* Tris (the incubation mixture), and 6.6×10^{-5} *M* Mg-ATP was added to initiate a reaction. The second half of the fiber bundle was incubated with the incubation mixture and the following test substances: 16 mg/ml PG, 33 mg/ml PG, 66 mg/ml PG, 33 mg/ml PPG 225, 66 mg/ml PPG 225, 33 mg/ml PPG 425, or 66 mg/ml PPG 425. All doses of PG, PPG 225, and PPG 425 promoted shortening of the test muscle fibers, as well as development of tension within the test fibers.

An aqueous solution of PPG 400 (dose 20 mg/kg) was administered intravenously to a dog (weight not stated) that was curarized, but not anesthetized. The results of an electroencephalogram (EEG) indicated increased electrical activity in all lobes of the brain. The increased activity was first noted in the frontoparietal region and spread gradually to temporal and occipital regions. Following intravenous administration of a PPG 750 aqueous solution and an aqueous solution of PPG 1200 (doses 10 mg/kg), the same EEG pattern was noted. However, the spread of electrical activity to other areas of the brain was more rapid. Electrocardiogram patterns following the administration of PPG 400 and PPG 750 indicated positive chronotropic responses. However, such responses were not observed after administration of PPG 1200 or PPG 2000 (Shideman and Procita, 1951).

The administration of aqueous PPG 400, PPG 750, or PPG 1200 to the anesthetized, noncurarized dog (weight not stated) at doses of 5 mg/kg caused an increase in respiratory rate and amplitude. Manifestations of increased central nervous system activity in the form of enhanced stretch reflexes, muscle tremors, and movements were also noted. At a higher dose, 25 mg/kg, convulsant activity was noted (Shideman and Procita, 1951).

The intravenous administration of 10- and 50-mg/kg doses of PPG 425, diluted 1:10 in saline solution, to groups of eight anesthetized, adult mongrel dogs (weights not stated) caused acceleration of cardiac and respiratory rates. Effects of the smaller dose on the electrocardiogram pattern ranged from a barely perceptible change to the presence of occasional ventricular extrasystoles. Intravenous doses of 50 mg/kg resulted in violent clonic convulsions and increased the heart rate to nearly double that of the control value; respiration was described as shallow and spasmodic. Doses of PPG 1025 (1 g/kg) and PPG 2025 (5 g/kg) produced no appreciable effect on the electrocardiogram within 15 min postdosing (Shaffer et al., 1951).

The effects of PG on cardiopulmonary function were studied by a number of investigators. An intravenous bolus injection (0.5 ml/kg) of 30% PG in sterile water caused a transient significant increase in pulmonary arterial pressure in a

group of eight mixed breed sheep (weights 20–28 kg). The peak mean pulmonary arterial pressure increased from 17 ± 1 to a peak of 35 ± 4 mm Hg ($p < 0.05$), and significant increases in pressure were not noted at 5 min postdosing. A similar finding was reported in lambs, but not in dogs or guinea pigs (Quinn et al., 1990).

Eichbaum and Yasaka (1976) studied the antiarrhythmic effects of solvents, including PG. Twenty rats and 15 dogs were anesthetized prior to having arrhythmias (monitored by electrocardiograph) induced via ouabain injections. The animals were then dosed intravenously with 0.2 ml/kg body wt 100% PG or 0.2 ml/kg body wt 70% PG. At each dose administered, PG produced either a temporary or a permanent change to a normal cardiac rhythm.

These authors also investigated the local and general anesthetic effects of PG in rats, dogs, and rabbits. Four rats were given periarticular injections of 0.1 ml 70% PG. No significant local anesthetic effect was noted in any of the test animals. Four rats, two rabbits, and two dogs were given intravenous injections of up to 4 ml/kg body wt 100% PG (specific doses not stated). There were no significant general anesthetic effects noted in any of the animals within 60 min after dosing.

Thomas et al. (1980) investigated the antibacterial properties of PG. Agar plates were inoculated with the test organisms *Staphylococcus aureus, Pseudomonas aeruginosa, Streptococcus faecalis,* and *Streptococcus pyogenes.* After a 24-h incubation period, a 1.5-cm hole was cut in the center of each agar plate. A 100% solution of PG was poured into the hole, and the zones of inhibition of the test organism's growth were measured on each plate. PG did not produce any zones of inhibition in any of the plates. PG did not have antiseptic properties in this test.

TOXICOLOGY

Acute Inhalation Toxicity

Five male albino rats (weights 213–238 g) were exposed for 7 h to vapor saturated with PPG 1200. Gross pathologic examinations were conducted on one animal at day 3 postexposure and on another animal at the end of the study. There were no signs of toxicity in animals dosed with vapor generated at room temperature. Two of the animals exposed to vapors generated at 100°C had lung rales on day 10 postexposure and one rat had a bloody discharge on day 17. Weight gain was noted in all animals. Emphysema and a moderate degree of hepatic and renal involvement were noted in the animal killed on day 3 and in the animal killed at the end of the study, respectively (FDA, 1992).

Acute Oral Toxicity

Propylene Glycol

Sax (1979) reported an acute oral LD_{50} of 21 g/kg body wt for PG in rats.

Five female Fischer strain rats were administered doses of PG to determine the oral LD_{50} of the compound. The 24-h LD_{50} of PG was 22.8 g/kg body wt. The 95% confidence interval was 20.5–25.3 g/kg. The lowest recorded 24-h lethal dose in this experiment was 20.9 g/kg, and the lowest recorded 48-h lethal dose was 19.8 g/kg (Clark et al., 1979).

Bartsch et al. (1976) reported an oral LD_{50} of 25 ml/kg in 10 male and female Sprague–Dawley rats. The 95% confidence level was 19.2–32.5 ml/kg.

Saini et al. (1987) investigated the effects of acute oral administration of PG using female Wistar strain rats. Three groups of rats were given 1-ml oral doses of 9.66, 19.32, or 38.64 mmol/kg body wt PG. Blood samples were drawn from the optical sinus at 0 h, 30 min, 60 min, 120 min, and 24 h postdosing. Acute oral administration of PG caused significant decreases in the levels of fibrinogen, albumin, and globulin in the plasma. PG affected the function of the liver in either the synthesis or the secretion of proteins in the rat.

The accidental oral administration of 3.8 L of PG to a male horse (weight 450–500 kg) resulted in pain, ataxia, salivation, and excessive sweating within 10–15 min. With the exception of ataxia, all clinical signs had resolved within 5 min, and an appropriate treatment protocol was begun. On the next day, the animal became increasingly ataxic and died of apparent respiratory arrest. Death occurred at 28 h postingestion. No appreciable gross lesions were noted at necropsy; PG concentrations of 9,000 mg/L in serum and 7,500 mg/L in renal fluids (combined urine and blood) were detected. Microscopic changes included moderate myocardial perivascular edema with dilatation of lymphatics and moderate pulmonary edema characterized by proteinaceous material in alveoli and in some of the bronchioles. Hepatic lesions consisted of scattered single-cell hepatocytic necrosis and minimal acute suppurative pericholangitis. Peracute renal infarcts characterized by multiple linear areas of coagulative tubular necrosis were also observed (Dorman and Haschek, 1991).

Polypropylene Glycols

The acute oral toxicity of PPGs (20% aqueous) was evaluated using three groups of five male albino rats of the Sherman strain (weights between 90 and 120 g). The mean LD_{50} values reported for the three groups were as follows: PPG 425 (2.91 g/kg), PPG 1025 (2.15 g/kg), and PPG 2025 (9.76 g/kg). Sluggishness, prostration, tremors, convulsions, and rapid death were noted in all treatment groups. The following gross lesions were observed in animals that died: minor pulmonary hemorrhage, congestion of the liver and spleen, renal ischemia, and injection of intestinal blood vessels (Shaffer et al., 1951).

In another study, 121 male rats (seven groups, 13–20/group) received various oral doses of 10% aqueous PPG 1200. The LD_{50} was 640 mg/kg (confidence limits 376–1,088 mg/kg) (FDA, 1992).

Additional acute oral toxicity studies on PPGs of various molecular weights (300–3,900) have indicated LD_{50} values (rats) ranging from 0.5 to >40 g/kg (American Industrial Hygiene Association, 1980).

An aqueous solution of PPG 1200 was tested in an acute oral toxicity study involving 10 dogs. Neither the test substance concentration nor the weight range of animals tested was stated. A subconvulsant effect was noted after the administration of 50 mg/kg (Shideman and Procita, 1951).

The acute oral toxicity of 50% aqueous PPG 1200 was evaluated using 55 male and 63 female guinea pigs (six groups/sex; 7–15/group). The LD_{50} values for male

and female guinea pigs were 1,320 mg/kg (95% confidence limits 930–1,875 mg/kg) and 1,420 mg/kg (95% confidence limits 1,090–1,850 mg/kg), respectively (FDA, 1992).

Acute oral toxicity studies on PPGs of various molecular weights have indicated LD_{50} values (guinea pigs) ranging from 1.5 to 17 g/kg (American Industrial Hygiene Association, 1980).

Acute Dermal Toxicity

The dermal toxicity of PPGs (undiluted) was evaluated according to the procedure of Draize et al. (1944) using groups of five rabbits. Doses of PPG 425 (10 and 20 ml/kg), PPG 1025 (20 ml/kg), and PPG 2025 (20 ml/kg) were applied for 24 h, and animals were observed for 13 days. All animals dosed with PPG 1025 and PPG 2025 survived the 24-h contact period and the observation period. Two of the five rabbits dosed with PPG 425 (20 ml/kg) and one of five rabbits dosed with 10 ml/kg PPG 425 died; animal deaths resulted from dermal penetration of the test substance. It was concluded that PPG 1025 and PPG 2025 did not penetrate the skin readily and that PPG 425 penetrated the skin to some extent (Shaffer et al., 1951).

In another study, the dermal toxicity of undiluted PPG 1200 was evaluated using 10 albino rabbits (weights 2.43–3.82 kg). The distribution of doses applied was as follows: 1.0 ml/kg body wt (two rabbits), 3.0 ml/kg (two rabbits), 10.0 ml/kg (one rabbit), and 30.0 ml/kg (five rabbits). Doses (24-h exposures) were applied under a heavy-gauge plastic cuff held in place with rubber bands and a cloth bandage that was taped securely to the marginal hair. At the end of exposure, sites were washed with soap and water, rinsed thoroughly, and dried with a cloth towel. The animals were observed for signs of toxicity during exposure and periodically for 2 weeks after exposure. Doses of 1.0, 3.0, or 10.0 ml/kg did not cause death, and, at most, moderate edema and slight erythema were observed at application sites. Body weight gains were noted in all animals, including the 30-ml/kg dose group. One of five rabbits in the 30.0-ml/kg group died, and the remaining rabbits appeared ill during the 2-week observation period. Overt signs of toxicity were not noted in these rabbits at the conclusion of the study (FDA, 1992).

Following single 24-h dermal applications of PPGs to animals (dose 30 ml/kg; species not stated), four of five animals survived treatment with PPG 400 or PPG 1200, and six of six animals survived treatment with PPG 200. All test substances were said to have been poorly absorbed through the skin (American Industrial Hygiene Association, 1980).

Acute Intraperitoneal Toxicity

Propylene Glycol

In the rat, the reported intraperitoneal LD_{50} for PG was 13 g/kg (Sax, 1979).

Bartsch et al. (1976) reported an intraperitoneal LD_{50} of 13.0 g/kg for PG in male and female Sprague–Dawley rats. The 95% confidence level was 10.2–16.5 ml/kg.

Six male NMRI mice were given intraperitoneal doses of PG diluted in water to

10 ml/kg body wt. The intraperitoneal LD_{50} was 11.2 g/kg body wt. Confidence levels were 11.0–11.5 g/kg (Budden et al., 1979).

Groups of five male and five female C3H strain mice were given intraperitoneal injections of 5 ml/kg PG. Mice were observed for 7 days. All had signs of lack of coordination followed by deep narcosis. There were no deaths during the 7-day observation period. No changes were noted in weight gain, feed and water consumption, or general behavior. At necropsy, peritonitis was observed in the test animals (Hickman, 1965).

Polypropylene Glycols

The acute intraperitoneal toxicity of PPGs (undiluted) was evaluated using three groups of five male albino rats of the Sherman strain (weights between 90 and 120 g). The mean LD_{50} values reported for the three groups were as follows: PPG 425 (0.46 g/kg), PPG 1025 (0.23 g/kg), and PPG 2025 (4.47 g/kg). In all treatment groups, death was preceded by tremors, prostration, frothing at the mouth, and audible rales (Shaffer et al., 1951).

Acute intraperitoneal LD_{50} values that have been reported for aqueous PPG solutions are as follows: PPG 400 (700 ± 75 mg/kg, 50 mice), PPG 750 (195 ± 8 mg/kg, 50 mice), PPG 1200 (113 ± 12 mg/kg, 60 mice), and PPG 2000 (3,600 ± 400 mg/kg). Weight ranges (not stated) were similar between all treatment groups (Shideman and Procita, 1951).

The results of other acute intraperitoneal toxicity studies have indicated LD_{50} values (rats) of 0.1–0.7 g/kg for PPGs ranging in molecular weight from 400 to 1,200 and an LD_{50} of 3.6 g/kg for PPG 2000 (American Industrial Hygiene Association, 1980).

Acute Intravenous Toxicity

Propylene Glycol

Bartsch et al. (1976) reported an intravenous LD_{50} for PG of 6.2 ml/kg in male and female Sprague–Dawley rats. The 95% confidence level was 5.2–7.4 ml/kg.

The same authors reported an intravenous LD_{50} of 6.4 ml/kg in male and female SPF-NMRI strain mice. The 95% confidence level was 6.1–6.9 ml/kg.

Polypropylene Glycols

The acute intravenous toxicity of PPGs (undiluted) was evaluated using three groups of five albino rats of the Sherman strain (weights between 90 and 120 g). The mean LD_{50} values reported for the three groups were as follows: PPG 425 (0.41 g/kg), PPG 1025 (0.12 g/kg), and PPG 2025 (0.71 g/kg). In all treatment groups, death was preceded by tremors, prostration, frothing at the mouth, and audible rales. Convulsions were noted only in groups treated with PPG 1025 and PPG 2025 (Shaffer et al., 1951).

Aqueous PPG solutions were tested in acute intravenous toxicity studies involving dogs (10 dogs/study; weights not stated). The results were as follows (Shideman and Procita, 1951): PPG 400 (dose 10–20 mg/kg, tremors with convul-

sions); PPG 750 (5–7 mg/kg, tremors), PPG 750 (8–15 mg/kg, dogs with convulsions survived); PPG 750 (20 mg/kg, dogs with convulsions died), PPG 1200 (5–7 mg/kg, tremors), PPG 1200 (15 mg/kg, dogs with convulsions survived), PPG 1200 (20 mg/kg, dogs with convulsions died), and PPG 2000 (100 mg/kg, no visible effects).

Acute Intramuscular Toxicity

The acute intramuscular toxicity of aqueous solutions of PPG 1200 was evaluated using two groups of 10 dogs (weights not stated). Subconvulsant effects were noted in dogs injected with 45 mg/kg PPG 1200, and mild convulsions were noted in dogs injected with 50–60 mg/kg PPG 1200 (Shideman and Procita, 1951). The results of acute toxicity studies on PG and PPG are summarized in Tables 7 and 8, respectively.

Short-Term Oral Toxicity

The effects of short-term administration of PG in rats and dogs were studied by Staples et al. (1967). Charles River rats (10/group) were given PG via stomach tube three times a day for 3 days. Doses were 0.75 ml/kg 100% PG, 1.50 ml/kg 100% PG, 3.0 ml/kg 100% PG, 3.0 ml/kg 75% PG, and 3.0 ml/kg 50% PG. Three adult mongrel dogs were also given PG via stomach tube for the same time period. Doses were 0.75, 1.5, or 3.0 ml/kg PG. One rat given the smallest dose and two rats given 3 ml/kg 100% PG had slight hyperemia of the GI tract. One rat given 1.5 ml/kg 100% PG had severe hyperemia of the GI tract. No other compound-related effects were noted in the rats. The dog given 1.5 ml/kg PG had evidence of patches of slight hyperemia. No other adverse effects were noted in any of the dogs.

The oral toxicity of PG was evaluated using a total of 11 male and female mongrel cats (weight range 3–4 kg) (Christopher et al., 1990*b*). Five cats were fed a diet containing 12% PG (dry weight) for 5 weeks; this diet was preceded by 4 weeks on a control diet containing 12% PG (dry weight). The remaining six cats were fed a diet containing 41% PG (dry weight) for 22 days. Mean values for

TABLE 7. *Reported LD_{50} values for Propylene Glycol*

Animal	Strain	Sex	Dose	Results	Ref.
Rat	—	—	Oral	21 g/kg	Sax (1979)
Rat	Fischer	F	Oral	22.8 g/kg	Clark et al. (1979)
Rat	Sprague–Dawley	M, F	Oral	25 ml/kg	Bartsch (1976)
Rat	—	—	Oral	27 g/kg	Layton et al. (1987)
Rat	—	—	i.p.	13 g/kg	Sax (1979)
Rat	Sprague–Dawley	M, F	i.p.	13 ml/kg	Bartsch (1976)
Rat	SPF	M, F	i.p.	13.7 ml/kg	Hickman (1965)
Mouse	NMRI	M	i.p.	11.2 g/kg	Budden et al. (1979)
Mouse	SPF–NMRI	M, F	i.p.	9.3 ml/kg	Bartsch (1976)
Rat	Sprague–Dawley	M, F	i.v.	6.2 ml/kg	Bartsch (1976)
Mouse	SPF–NMRI	M, F	i.v.	6.4 ml/kg	Bartsch (1976)
Mouse	—	—	i.v.	8.0 g/kg	Sax (1979)
Rat	—	—	i.m.	20.0 g/kg	Sax (1979)
Mouse	—	—	s.c.	18.5 g/kg	Sax (1979)

TABLE 8. *Acute toxicity studies on Polypropylene Glycol (PPG)*

Animal	Strain	Sex	Dose	Results	Ref.
Rat	Sherman	M	Oral	PPG 425: LD_{50} = 2.91 g/kg	Shaffer et al. (1951)
Rat	Sherman	M	Oral	PPG 1025: LD_{50} = 2.15 g/kg	Shaffer et al. (1951)
Rat	—	M	Oral	PPG 1200: LD_{50} = 640 mg/kg	FDA (1992)
Rat	Sherman	M	Oral	PPG 2025: LD_{50} = 9.76 g/kg	Shaffer et al. (1951)
Dog	—	—	Oral	Subconvulsant effect at 50 mg/kg	Shideman and Procita (1951)
Guinea pig	—	M, F	Oral	PPG 1200: LD_{50} = 1,320 mg/kg (males) and 1,420 mg/kg (females)	American Industrial Hygiene Association (1980)
Rabbit	—	—	Dermal	PPG 425: 20% and 40% mortality at 10 and 20 ml/kg	Shaffer et al. (1951)
Rabbit	—	—	Dermal	PPG 1025: no deaths at 20 ml/kg	Shaffer et al. (1951)
Rabbit	—	—	Dermal	PPG 1200: 20% mortality at 30 ml/kg	FDA (1992)
Rabbit	—	—	Dermal	PPG 2025: no deaths at 20 ml/kg	Shaffer et al. (1951)
Mouse	—	—	i.p.	PPG 400: LD_{50} = 700 mg/kg	Shideman and Procita (1951)
Mouse	—	—	i.p.	PPG 750: LD_{50} = 195 mg/kg	Shideman and Procita (1951)
Mouse	—	—	i.p.	PPG 1200: LD_{50} = 113 mg/kg	Shideman and Procita (1951)
Mouse	—	—	i.p.	PPG 2000: LD_{50} = 3,600 mg/kg	Shideman and Procita (1951)
Rat	Sherman	M	i.p.	PPG 425: LD_{50} = 0.46 g/kg	Shaffer et al. (1951)
Rat	Sherman	M	i.p.	PPG 1025: LD_{50} = 0.23 g/kg	Shaffer et al. (1951)
Rat	Sherman	M	i.p.	PPG 2025: LD_{50} = 4.47 g/kg	Shaffer et al. (1951)
Rat	Sherman	—	i.v.	PPG 425: LD_{50} = 0.41 g/kg	Shaffer et al. (1951)
Rat	Sherman	—	i.v.	PPG 1025: LD_{50} = 0.12 g/kg	Shaffer et al. (1951)
Rat	Sherman	—	i.v.	PPG 2025: LD_{50} = 0.71 g/kg	Shaffer et al. (1951)
Dog	—	—	i.v.	PPG 400: tremors with convulsions, at 10–20 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 750: tremors at 5–7 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 750: convulsions but no deaths at 8–15 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 750: convulsions and deaths at 20 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 1200: tremors at 5–7 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 1200: convulsions but no deaths at 15 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 1200: convulsions and deaths at 20 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.v.	PPG 2000: no visible effects	Shideman and Procita (1951)
Dog	—	—	i.m.	PPG 1200: subconvulsant effects at 45 mg/kg	Shideman and Procita (1951)
Dog	—	—	i.m.	PPG 1200: mild convulsions at 50–60 mg/kg	Shideman and Procita (1951)

ingested PG were 1.6 g/kg body wt/day for the 12% dose group and 8.0 g/kg body wt/day for the 41% dose group. In the low-dose group, plasma D-lactate (plasma samples obtained only from two cats) increased from 0.08 ± 0.03 mmol/L (range 0–0.16 mmol/L) during the control period to 1.96 ± 0.75 mmol/L (range 0.2–4.2 mmol/L) on day 24. In the high-dose group, plasma D-lactate increased rapidly to 4.21 ± 1.96 mmol/L by day 10 ($p < 0.001$), and on day 24 a mean value of 7.12 ± 0.14 mmol/L was reported. These findings indicate that PG is metabolized in vivo, at least in part, to D-lactate. The results of physical examinations performed on cats that received the smaller dose did not indicate any abnormal findings. Observations in cats that received the high dose included moderate polyuria and polydypsia; these findings are consistent with renal excretion of PG, which acts as an osmotic diuretic (Hanzlik et al., 1939). Cats in the high-dose group also developed decreased activity, mental depression, and slight to moderate ataxia. These observations may be related to the metabolism of PG to D-lactate. For example, encephalopathy in humans, characterized by disorientation and ataxia, is associated with D-lactate concentrations of >4.0 mmol/L (Ludvigsen et al., 1983).

Short-Term Intravenous Toxicity

Fort et al. (1984) investigated the effects of short-term administration of PG using rats and dogs. Groups of five male and five female rats were given intravenous infusions of PG/ethanol/water (5:1:4) over a period of 2 weeks. The dose was 5 ml/kg/day at a rate of 0.3 ml/min. One male beagle and one female beagle dog were given the same PG/ethanol/water mixture at doses of 4 ml/kg/day at a rate of 6 ml/min. Excretion of red urine was noted after the initial dose in both test species and continued throughout the 2-week study in most rats and in some dogs (number not specified). After the 2-week treatment period, decreases in packed cell volume, hemoglobin, and erythrocyte counts were noted in both test species. At necropsy, splenomegaly and renal hemosiderin deposits were observed in the rats.

Subchronic Oral Toxicity

Propylene Glycol

Gaunt et al. (1972) investigated the effects of subchronic administration of PG using rats. Fifteen male and 15 female Charles River CD rats were fed diets containing 0 (control) or 50,000 ppm PG for 15 weeks. The PG administered in the feed corresponded to a dose of 2.5 g/kg/day in the test animals. Blood samples were collected at the end of the study, and a hematological examination was performed. The brain, heart, liver, spleen, kidneys, adrenal glands, gonads, and pituitary gland were collected from each animal, and organ weights were compared with those of controls. No significant differences were found between the control and PG-dosed animals. No compound-related lesions were noted.

Polypropylene Glycols

PPG 2000 was administered to rats (weights, strain, and number tested not stated) over a period of 100 days. Concentrations of 0.1, 0.3, 1.0, and 3.0% were

administered in oral doses of ~50–1,500 mg/kg/day. Adverse effects were not noted at concentrations of 0.1–1.0%. Slight decreases in growth were observed after the administration of 3% PPG 2000 (American Industrial Hygiene Association, 1980).

In a 90-day study, PPG 2000 was administered orally to rats (weights and number tested not stated) in doses ranging from 275 to 501 mg/kg/day. There was no evidence of adverse histopathologic, hematologic, or clinical chemistry effects in any of the animals tested. Body weight effects (not specified) were noted at the highest dose tested. Similar results were reported for dogs (weights and number tested not stated) that received doses of PPG 2000 ranging from 526 to 810 mg/kg/day (American Industrial Hygiene Association, 1980).

In another study, PPG 1200 was fed to three groups of four purebred beagle dogs (weights 5.95–12.5 kg) for 90 days. Each group consisted of two male and two female dogs. The three groups ingested PPG 1200 at concentrations of 0.1, 0.3, and 1.0% in the diet, respectively; six beagles in the control group were fed diets without PPG. At the highest "no effect level" (0.3%), average daily consumption doses were 79 and 99 mg/kg for two male dogs and 90 and 123 mg/kg for two female dogs. Data based on the following analyses did not indicate adverse effects at any of the concentrations tested: demeanor, feed consumption, hematologic parameters, clinical chemistry determinations, urinalysis, organ weights, organ/body weight and organ/brain weight ratios, or gross and microscopic examination of tissues and organs. Similar results were reported when three groups of 50 rats were dosed with 0.1, 0.3, and 1.0% PPG 1200, respectively, according to the same procedure. Each group was equally mixed and weight ranges for male and female rats were 321–334 and 218–233 g, respectively (FDA, 1992).

PPG 750 was administered to rats (weights and number tested not stated) over a period of 100 days. Concentrations of 0.1 and 1% were administered at doses of ~50 and 500 mg/kg/day. PPG 750 (0.1%) did not induce any adverse effects. However, in the group dosed with 1% PPG 750, there was a slight increase in liver and kidney weights; there were no histological changes. Neither of the doses resulted in a central nervous system stimulatory effect (American Industrial Hygiene Association, 1980).

Subchronic Dermal Toxicity

Propylene Glycol

PG (0.2 ml) was injected subcutaneously into 12 male hairless mice of the *hr/hr* Oslo strain (10–12 weeks old) three times per week for 3 months. None of the animals died and there was no ulceration at the injection sites. Based on microflow histograms of urinary bladder epithelial cells (Farsund, 1974), the following results were reported: The proportion of diploid cells in the S-phase fluctuated but was not significantly altered. The proportion of tetraploid cells in the S-phase was significantly reduced, and at 3 months, there was no DNA synthesis in these cells. Overall, the proportion of diploid cells increased, the number of tetraploids was slightly reduced, and almost all of the octoploid cells disappeared. The alterations reported were described as disturbed regenerative reactions resulting from PG-

induced acute cellular toxicity. Some of the bladder epithelial cells were killed and the mechanism of repeated DNA synthesis was disturbed (Farsund, 1978).

Polypropylene Glycol

In a 3-month study, PPG 2000 was applied to the skin of rabbits (weights and number tested not stated) at doses of 1, 5, and 10 ml/kg. Applications were made 5 days per week (24 h/day). Doses of 1 ml/kg did not cause any adverse effects. Slight depression of growth was noted after the administration of 5- and 10-ml/kg doses (American Industrial Hygiene Association, 1980).

Chronic Oral Toxicity

In a chronic feeding study by Gaunt et al. (1972), PG was administered in the diet to rats for 2 years. Groups of 30 male and female Charles River CD rats were fed diets containing 0, 6,250, 12,500, 25,000, or 50,000 ppm PG. Feed and water were given ad libitum. General behavior was observed, and body weights were recorded every 2 weeks. At 104 weeks, animals were killed and brain, heart, liver, spleen, kidneys, adrenal glands, stomach, small intestines, and cecum were weighed. Samples of organs that appeared to have lesions were retained for microscopic examination. No significant compound-related lesions were noted in the test animals; all organ and body weights in the treated animals were not significantly different from controls.

Weil et al. (1971) investigated the effects of chronic administration of PG using dogs. Groups of five male and five female dogs were given diets containing 2 to 5 g/kg PG. Body weights were recorded throughout the experiment. At week 104, all animals were killed and organs were examined microscopically for lesions. At the 5-g/kg dose levels, dogs gained more weight than controls, and urine output was increased. No compound-related lesions were noted. The no-effect level for PG was 2 g/kg body wt.

A group of four female dogs (weights 6–21 g) received 5% PG (in drinking water) for a period ranging from 5 to 9 months. The animals were allowed to drink the mixture twice daily during a period of 1 h. Additionally, each of four male dogs (weights not stated) was allowed to drink 600 ml of 10% PG (in drinking water) daily over a period ranging from 5 to 6 months. The three tests of liver function included galactose excretion, uric acid excretion, and the rose bengal content of blood. The excretion of phenolsulfonphthalein during a 2-h test period served as the test for renal function. The results indicated no impairment of hepatic or renal function in either male or female dogs (Van Winkle and Newman, 1941).

Ocular Irritation

Propylene Glycol

Clark et al. (1979) evaluated the ocular irritation potential of solar heat transfer fluids, including PG. PG (0.1 ml) was instilled into the conjunctival sac of one eye of each of six rabbits, and reactions were scored according to the methods of Draize et al. (1944). Eyes were examined at 1, 24, 48, 72, and 96 h posttreatment.

At 24 h, a score of 0.7 (maximum score = 110) was noted. At 72 h, the score was 0.3. At 96 h, the score was 0.0. There was no corneal damage noted in any of the test animals.

The ocular irritation potential of 56 substances, including PG, was investigated by Guillot et al. (1982*a*). PG (pH 8.8; 0.1 ml) was instilled into the conjunctival sac of one eye of each of six male New Zealand White rabbits. Untreated eyes served as controls. Eyes were not rinsed after instillation. The acute ocular irritation index (AOI) (maximum possible score = 110) and the mean ocular irritation index (MOI) (maximum possible score = 40) were calculated for PG. The AOI was 11.33 and the MOI was 0.83. PG was a slight ocular irritant in this test.

Douglas and Spilman (1983) conducted an in vitro ocular irritancy test using several substances, including PG. A ^{51}Cr release assay using a cultured corneal endothelial cell line was used to approximate ocular toxicity in vivo. Cells were preincubated with 0.5 μCi/ml ^{51}Cr for 1 h. The corneal endothelial cells were then incubated with the test substance. Different molar concentrations of the test substance were used to obtain the estimated dose that resulted in 50% toxicity (ED_{50}). The test was run three times, and the ED_{50} values measured were 30, 13, and 13 *M*. The 95% confidence limits were 15–61, 7.2–24, and 8.2–20 *M*, respectively. PG was a nonirritant in this test.

Polypropylene Glycols

Following instillation of an excess of PPG 425, PPG 1025, and PPG 2025 into the conjunctival sacs of rabbits, trace injuries to one or two of five eyes tested with each test substance were observed. PPG 425, PPG 1025, and PPG 2025 were classified as harmless agents (Shaffer et al., 1951).

The ocular irritation potential of undiluted PPG 1200 was evaluated using an albino rabbit. The test substance (0.1 ml) was instilled into the conjunctival sac of both eyes; only one eye was rinsed after instillation. Both eyes were examined for conjunctival irritation, corneal injury, and internal effects such as iritis and lenticular damage. At most, the test substance induced very slight discomfort and slight transient conjunctival irritation. Signs of ocular irritation had cleared by 24 h postinstillation (FDA, 1992).

Skin Irritation/Sensitization

Propylene Glycol

The skin irritancy potential of PG was studied by Lashmar et al. (1989) using nude mice. PG (10, 25, or 50%) was instilled in polyvinyl chloride cups (vol 0.3 cm^2) on the dorsal side of three mice. The test substance remained in contact with the skin for 24 h. At the end of 24 h, the animals were killed and a sample of the exposed skin was taken for microscopic examination. Reactions were scored according to the methods of Ingram and Grasso (1975) (maximum possible score = 77). At 10%, no irritation was observed. At 25%, the score was 5, and at 50%, the score was 11. In the animals exposed to 50% PG, hypertrophy, dermal in-

flammation, and proliferation stimuli were noted, indicating that this concentration of PG may cause skin irritation.

In a mouse external ear sensitization assay, 100% PG was applied to the right ear of each of 19 mice (Swiss, BALB-c, CBA, C56B1/6, DBA-2, and B6D2F1 strains) on days 0 and 2 of the experiment. On day 2, Freund's Complete Adjuvant was injected subcutaneously into the external ears of each mouse. On day 9, the thickness of the external ear was measured with a mobile-disk caliper immediately before and 24 h after a topical application of 100% PG to the treated external ears of each of the test mice. There was no significant increase in the external ear thickness, a measure of skin irritation and sensitization, in any of the test animals as compared with controls (Descotes, 1988).

Clark et al. (1979) investigated the irritant effects of PG in rabbits. PG (concentration not stated) was applied to intact and abraded skin on the backs of six New Zealand white rabbits according to the methods of Draize et al. (1944). Irritation was scored at 24 and 72 h after treatment. The irritation score, obtained by a compilation of scores obtained at 24 and 72 h, was 0.1 (maximum possible = 8). A score of <2 was classified as a mild to no irritation reaction.

PG was applied to the clipped skin (intact and scarified) of the backs of three groups of six New Zealand white rabbits according to three different protocols by Guillot et al. (1982*b*). In protocol 1 (the cosmetic protocol), rabbits were exposed to 0.5 ml of the test substance for 23 h under occlusive patches. Protocol 2 [the Association Francaise de Normalisation (AFNOR) protocol] was defined as the exposure of rabbits to 0.5 ml of the test substance for 4 h under occlusive patches. In protocol 3 [the Organisation for Economic Co-operation and Development (OECD) protocol], rabbits were exposed to 0.5 ml of the test substance for 4 h under semiocclusive patches. In each of the three protocols, both abraded and intact skin sites were tested. Reactions were scored at 1 and 48 h in the cosmetic protocol, 1, 24, and 48 h in the AFNOR protocol, and 30–60 min, 24, 48, and 72 h in the OECD protocol. The primary cutaneous irritation index obtained for the three protocols were 0.17, 0.14, and 0.02, respectively. In all three tests, PG was classified as a nonirritant.

Open and closed patch tests were conducted by Motoyoshi et al. (1984) using guinea pigs and rabbits. Patches containing 0.1 ml PG/patch (four per animal; two open, two closed) were applied for 48 h to the shaved skin of six Hartley guinea pigs. Patches containing 0.3 ml PG/patch (four per animal; two open, two closed) were applied to the shaved skin of six Angora rabbits for 48 h. Reactions were scored 30 min after removal of the patches. There were no reactions noted in either the open or the closed patch tests in either the rabbits or the guinea pigs. The authors noted that both of the species tested lack sweat glands, which might have accounted for the lack of a reaction.

Forty-eight-hour and 21-day open and closed patch tests were conducted by these authors using miniature Gottingen swine. Four patches per animal (two open, two closed) containing 0.1 ml PG/patch were applied to the shaved skin of two swine. Reactions were scored 30 min after the 48-h exposure. In the 21-day test, reactions were scored daily. Skin irritation reactions were not observed in any of the animals. The authors concluded that the previously mentioned lack

of sweat glands may have contributed to the lack of a reaction after exposure to PG.

Pure PG was applied to the flanks of 27 guinea pigs and to the backs of 20 rabbits in a skin fold thickness test. Open patches containing the test material (amount unspecified) were applied daily to the test sites. Untreated sites served as controls. The skin fold thickness, a measurement of skin irritation, was measured daily with a Harpenden skin fold caliper. Sites were also examined for erythema, edema, and scaling. There were no significant increases in skin fold thickness in the rabbits. Most test sites in the rabbits had transient redness, which resolved spontaneously. There was a significant increase in skin fold thickness in the guinea pigs on day 7 ($p < 0.01$ and on days 8–10 ($p < 0.001$) (Wahlberg and Nilsson, 1984).

Kero and Hannuksela (1980) tested the induction of contact hypersensitivity reactions by PG using three different tests. Using the guinea pig maximization test (GPMT), 20 animals were injected with 70% PG. In the open epicutaneous test (OET), 20 guinea pigs had open patches containing 70% PG applied to an 8-cm^2 shaved area of skin during a 21-day period. In the chamber test (CT), eight Finn chambers containing 20 μl 70% PG were applied to the shaved skin of each of 20 guinea pigs for 6 days. Chambers were changed every 48 h. Challenges were done on day 21 in the GPMT and the CT and on day 31 in the OET. At challenge, no positive reactions were noted in any of the animals in the three tests. PG did not induce contact hypersensitivity in these tests.

Using the epicutaneous maximization test, Guillot and Gonnet (1985) investigated the sensitization potential of PG in guinea pigs. Seven successive 48-h occlusive patches containing PG (amount not specified) were applied to the shaved skin of each of 20 Dunkin–Hartley guinea pigs. After a 1-week nontreatment period, one 48-h occlusive challenge patch was applied to a previously untreated site on each guinea pig. Readings were taken at 1, 6, 24, and 48 h. Skin samples were taken for microscopic examination. No irritation or sensitization reactions were noted in any of the test animals. PG was neither an irritant nor a sensitizer in this test.

Guillot et al. (1983) studied the sensitizing potential of several chemicals, including PG, using seven different protocols. In the first protocol, the authors followed the methods of Magnusson and Kligman (1969) in a GPMT. On day 0 of the experiment, a 48-h occlusive patch containing 0.5 ml PG was applied to the dorsal surface of each of 20 Dunkin–Hartley guinea pigs. On day 7, the skin was painted with 0.5 ml 10% sodium lauryl sulfate. On day 8, a 48-h occlusive patch containing 0.5 ml PG was applied to the same area. After a 10-day nontreatment period, a 48-h occlusive challenge patch containing 0.5 ml PG was applied to the left flank of each animal. Reactions were scored 24 and 48 h after removal of the challenge patch. Skin samples were taken for microscopic examination. No reactions were noted in this test. PG was classified as a Grade I chemical in this test (Grade I = weak sensitizing potential).

In the second protocol, the split adjuvant technique, the authors followed the methods of Maguire (1973). Three groups of guinea pigs, 10 per group (5 males and 5 females), had 0.1 ml PG applied under an occlusive patch after a 5-s treatment

with dry ice on the dorsal surface (day 0 of the experiment). On day 2, a second 48-h occlusive patch was applied to the same area of the body. On day 4, animals received intradermal injections of 0.1 ml Freund's Complete Adjuvant, followed by a third 48-h occlusive patch containing 0.1 ml PG. After a 12-day nontreatment period, all groups had 24-h-occlusive patches containing 0.1 ml PG applied to the test areas on day 21 of the experiment. Group 1 animals were examined at 24, 48, and 72 h for reactions. Groups 2 and 3 received 24-h-occlusive patches containing the same amount of PG on day 35 of the experiment. Group 2 animals were examined at 24, 48, and 72 h for reactions. Group 3 animals received 24-h occlusive patches containing 0.1 ml PG on day 42 of the experiment and were examined 24, 48, and 72 h later for reactions. Results were scored on a five-point scale. PG was nonallergenic in this test.

Guillot et al. (1983) followed the methods of Maurer et al. (1975) to test the sensitization potential of PG in a guinea pig optimization test. Two intradermal injections of 0.1 ml PG were administered to each of 20 guinea pigs (10 males and 10 females) on day 0 of the experiment. On days 2 and 4, single injections of the same concentration of PG were administered, followed by depilation of the test site 21 h later. Reactions at the test sites were scored after depilation. On days 7, 9, 11 14, 16, and 18, animals received single intradermal injections of 50% PG/50% Freund's Complete Adjuvant. Following a 2-week nontreatment period, animals were depilated and received one intradermal injection of 0.1 ml PG on day 35. Test site reactions were scored after depilation. On day 49, 24-h occlusive patches containing 0.1 ml PG were applied to the test sites. On day 51, following the removal of the patches and depilation of the test site, irritation/sensitization reactions were evaluated. No reactions were noted in any of the test animals. PG was nonallergenic in this test.

These authors also investigated the irritation/sensitization potential of PG using a method outlined by Guillot and Brulos (unpublished protocol). In the induction phase of the experiment, 20 guinea pigs (10 male and 10 female) were each given single intradermal injections of 0.1 ml Freund's Complete Adjuvant, followed by 48-h occlusive patches containing 0.5 ml PG. On days 2, 4, 7, 9, 11, and 14, 49-h occlusive patches were reapplied. Following a 22-day nontreatment period, 48-h occlusive patches containing 0.5 ml PG were applied in the challenge phase of the experiment. At 1, 6, 24, and 48 h after removal of the challenge patches, macroscopic and histological examinations of the test sites were conducted. There were no reactions in any of the test animals, and the test score was 0 (0 = sensitizing potential weak or nonsignificant).

In the fifth protocol (Freund's Complete Adjuvant test), Guillot et al. (1983) followed the methods of Klecak et al. (1977). Two groups of 10 Dunkin–Hartley guinea pigs (5 male/5 female per group) received intradermal injections of 0.1 ml PG on days 0, 2, 4, 7, and 9 of the induction phase of the experiment. Following a 10-day nontreatment period, both groups received topical applications of PG (25 μl), and reactions were scored at 24, 48, and 72 h. Group 2 animals received an additional challenge of 25 μl PG on day 35, and reactions were scored at 24, 48, and 72 h. No reactions were noted. PG was nonallergenic in this test.

In the sixth test protocol, they followed the methods of Dossou and Sicard

(1984). Two groups of guinea pigs, 12 per group, were used to test the irritation/sensitization potential of PG. Group 1 animals received 0.5-ml dermal applications of PG in an open test on days 0, 2, and 4 of the experiment. Following a 10-day nontreatment period, challenge applications of 10 μl PG were applied to the skin. Reactions were scored 24 and 48 h after the challenge applications. Group 2 animals received injections of 50% PG/50% Freund's Complete Adjuvant on day 4 of the experiment. Following a 5-day nontreatment period, challenge applications of 10 μl PG were made to the skin. Reactions were scored at 24 and 48 h. No reactions were noted in either of the test groups. PG was nonallergenic in this test.

In the last protocol tested by Guillot et al. (1983), an OET was performed according to the methods of Klecak et al. (1977). Two groups of guinea pigs, eight per group, received single topical applications of 0.1 ml PG on the right posterior flank on days 0–20 of the experiment. On day 21, challenge applications of 25 μl PG were made to the test sites. Group 1 animals were examined at 24, 48, and 72 h for reactions. Following a 14-day nontreatment period, group 2 animals received additional challenge exposures of 25 μl PG per animal. Macroscopic and histological examinations were performed 24, 48, and 72 h later. No reactions were noted in any of the animals in either test group. PG was nonallergenic in this test.

Jacaruso et al. (1985) investigated the irritation potential of PG in an in vitro histamine release study. Cells were harvested from male Wistar rats, and a population containing mast cells, macrophages, leukocytes, and erythrocytes was isolated and suspended in HBSS and bovine serum albumin (BSA). PG, at final concentrations of 0.01, 0.10, 1.0, or 10%, was added to the cells for 10 min (37°C). The reaction mixture was spun at 400 *g* for 10 min, and the supernatant was used for analysis of histamine release, an index of irritation potential. Control values for histamine release were obtained by exposing a cell population to HBSS/BSA alone. Only those cells exposed to a final concentration of 1.0% PG released significantly more histamine than control cells ($p < 0.01$).

Polypropylene Glycols

Skin irritation was not noted after PPG 425, PPG 1025, or PPG 2025 when applied once to clipped skin (abdomen) of the rabbit or when applied a total of eight times to the same area within 4 h. Details concerning the experimental procedure were not stated (Shaffer et al., 1951).

The skin irritation potential of undiluted PPG 1200 was evaluated using an albino rabbit (weight 2.3 kg). The test substance (1–2 ml) was applied to intact and abraded abdominal skin by means of an absorbent cotton pad held in place by a cloth bandage that was taped to the trunk. Prolonged applications were made to intact skin (daily for 3 days) and abraded skin (5 days/week for 2 weeks). Repeated applications (5 days/week for 2 weeks) were also made to the inner surface of the external ear. Reactions were not observed at the intact skin site. Barely perceptible erythema and slight exfoliation were observed at the abraded site. There was no perceivable skin damage after multiple applications to the inner surface of the external ear (FDA, 1992).

The results of skin irritation and sensitization studies on PG are summarized in Table 9. Studies on the skin irritation potential of PPG are summarized in Table 10.

Hematotoxicity

The hemolytic effects of PG were evaluated in a 117-day study using 21 adult, specific-pathogen-free cats (7 males, 14 females; weights between 2.3 and 5.1 kg). The animals were randomly divided into three groups and fed 6% PG, 12% PG (in

TABLE 9. *Irritation/sensitization potential of Propylene Glycol*

Animal	No.	Dose/concentration; exposure method	Results	Ref.
Nude mice	3	10, 25, 50%; 24-h exposure in PVC cup	No irritation	Lashmar et al. (1989)
Mice	19	100%; mouse ear sensitization assay	No irritation/ sensitization	Descotes (1988)
Rabbits	6	Unknown concentration; 24-h patch test	Mild to no irritation	Clark et al. (1979)
Rabbits	6	Cosmetic protocol (see text)	Nonirritant	Guillot et al. (1982*b*)
Rabbits	6	AFNOR protocol (see text)	Nonirritant	Guillot et al. (1982*b*)
Rabbits	6	OECD protocol (see text)	Nonirritant	Guillot et al. (1982*b*)
Guinea pigs	6	0.1 ml; 48-h open patch test	No observed reaction	Motoyoshi et al. (1984)
Guinea pigs	6	0.1 ml; 48-h closed patch test	No observed reaction	Motoyoshi et al. (1984)
Rabbits	6	0.1 ml; 48-h open patch test	No observed reaction	Motoyoshi et al. (1984)
Rabbits	6	0.1 ml; 48-h closed patch test	No observed reaction	Motoyoshi et al. (1984)
Swine	2	0.1 ml; 48-h open patch test	No skin irritation	Motoyoshi et al. (1984)
Swine	2	0.1 ml; 48-h closed patch test	No skin irritation	Motoyoshi et al. (1984)
Swine	2	0.1 ml; 21-day open patch test	No skin irritation	Motoyoshi et al. (1984)
Swine	2	0.1 ml; 21-day closed patch test	No skin irritation	Motoyoshi et al. (1984)
Guinea pigs	27	Daily open patches, skin fold thickness assay	Days 7 and 8–10, significant increase in skin fold thickness	Wahlberg and Nilsson (1984)
Rabbits	20	Daily open patches, skin fold thickness assay	No increase in skinfold thickness; transient redness	Wahlberg and Nilsson, 1984
Guinea pigs	20	70%; maximization test	No observed reaction	Kero and Hannuksela (1980)
Guinea pigs	20	70%; open epicutaneous test	No observed reaction	Kero and Hannuksela (1980)
Guinea pigs	20	20 μl 70%; three 48-h chamber exposures	No observed reaction	Kero and Hannuksela (1980)
Guinea pigs	20	Epicutaneous maximization test	No irritation/ sensitization	Guillot and Gonnet (1985)
Guinea pigs	20	0.1 ml; protocol 1, maximization test	Weak irritation/ sensitization potential	Guillot et al. (1983)
Guinea pigs	30	0.1 ml; protocol 2, split adjuvant technique	Nonallergic	Guillot et al. (1983)
Guinea pigs	20	0.1 ml; protocol 3, optimization test	Nonallergic	Guillot et al. (1983)
Guinea pigs	20	0.5 ml; protocol 4, Guillot/Brulos	Nonsignificant or weak sensitizer	Guillot et al. (1983)
Guinea pigs	20	0.1 ml; protocol 5, Freund's Adjuvant test	Nonallergenic	Guillot et al. (1983)
Guinea pigs	24	Protocol 6 (see text)	Nonallergenic	Guillot et al. (1983)
Guinea pigs	16	0.1 ml; protocol 7, open epicutaneous test	Nonallergenic	Guillot et al. (1983)
Rats	—	0.01, 0.10, 1.0, and 10%; in vitro histamine release assay	Low skin irritation potential	Jacaruso et al. (1985)

For abbreviations and details, see the text.

TABLE 10. *Skin irritation potential of Polypropylene Glycol*

Animals	No.	Dose/concentration; exposure method	Results	Ref.
Rabbits	—	Single application and 8 applications within 4 h	No skin irritation	Shaffer et al. (1951)
Rabbits	—	Single application and 8 applications within 4 h	No skin irritation	Shaffer et al. (1951)
Rabbits	—	Single application and 8 applications within 4 h	No skin irritation	Shaffer et al. (1951)
Rabbit	1	Applications to intact skin daily for 3 days; applications to abraded skin 5 days/wk for 2 wks	No reactions at intact site; barely perceptible erythema and slight exfoliation at abraded site	FDA (1992)
Rabbit	1	Applications to different intact skin site 5 days/wk for 2 wks	No perceivable skin damage	FDA (1992)

the diet), and a control diet, respectively. Blood was drawn at the beginning of the study and every 2 weeks thereafter. Reticulocyte and Heinz body counts were expressed as the percentage of 1,000 cells counted. Differences between treatment groups and changes over time within groups were evaluated by an analysis of variance. In experimental and control groups, the packed cell volume decreased over time and was significantly lower in the 12% group only during the sixth week of the study. Significant changes in hemoglobin concentration were not observed between groups; however, a statistically significant ($p < 0.001$) decrease over time was observed in the 6% PG group. Throughout the study, a significant decrease ($p < 0.001$) in numbers of erythrocytes was noted in 6 and 12% PG groups, but not in the control group. A significant ($p < 0.0001$) increase in punctate reticulocytes over time was observed in 6 and 12% PG groups, but not in the control group. Additionally, a significant increase in the number of aggregate reticulocytes was noted in the 12% PG group 2 weeks after initiation of PG-containing diets. Heinz body counts were significantly higher, in a dose-related manner, in 6 and 12% PG groups; the increase persisted throughout the study. The results of this study indicated that PG-containing diets caused a dose-dependent erythrocyte destruction, even when fed at concentrations as low as 6% (Bauer et al., 1992). Decreased erythrocyte survival and Heinz body formation were noted in an earlier study in which three cats (weights 3–4 kg) were fed a dosage of 8 g PG/kg body wt daily for 22 days (Christopher et al., 1990*b*). Heinz body formation was also noted in groups of six kittens (weight 2,160 ± 47 g) fed diets containing 5 and 10% PG for 12 weeks. None of the animals had anemia or methemoglobinemia (Hickman et al., 1990).

Reproductive Effects

A continuous breeding reproduction study was conducted using COBS Crl:CD-1 (ICR)BR outbred Swiss albino mice (6 weeks old). The continuous breeding phase of the study (task II) was begun after the dose-setting study (task

I) and involved three experimental groups (40 mice per group) and a control group of 80 mice. Experimental and control groups contained an equal number of male and female mice. The three experimental groups were given the following doses (in feed or water), respectively, during a 7-day premating period: 1.0% PG (daily dose 1.82 g/kg), 2.5% PG (daily dose 4.80 g/kg), and 5.0% PG (daily dose 10.10 g/kg). The animals were then randomly grouped as mating pairs, cohabitated, and treated continuously for 98 days; data were collected on all newborns. If significant adverse effects on fertility were observed, a crossover mating trial (task III) was usually performed to determine whether F_0 males or females were more sensitive to the effects. Task III was not conducted and would have consisted of mating high-dose mice of each sex to control mice of the opposite sex and then analyzing the offspring. To perform an offspring assessment of reproductive function (task IV) following exposure to PG, the dam (from phase II) was dosed through weaning and F_1 mice were dosed until mating occurred at 74 ± 10 days of age. Mating pairs consisted of male and female offspring from the same treatment group (20/group/sex); F_2 litters were examined. In the continuous breeding phase (task II), there were no significant changes ($p < 0.05$) in mean live pup weight per litter between the control group and any of the treatment groups. In task IV, which was an offspring assessment of reproductive function, only the high-dose group (5% PG) was involved. There were no significant differences ($p < 0.05$) between control and experimental groups with respect to the following observations in task IV: mating index, fertility index, mean number of live pups per litter, proportion of pups born alive, and sex of pups born alive (Morrissey et al., 1989).

Embryotoxicity

The effect of PG on the development of $B_6D_2F_1$ mouse zygotes in the pronuclear stage was evaluated; oocytes were fertilized in vitro. Samples of zygotes were incubated for 20 min (at 22°C) with 1.5, 3.0, and 6.0 *M* PG in phosphate-buffered saline. There were three zygote cultures per test concentration of PG, and each group of three were incubated with 0, 0.1, and 0.25 *M* sucrose, respectively. The three control cultures without PG were also incubated with 0, 0.1, and 0.25 *M* sucrose, respectively. Subsequently, the zygotes were incubated for 24 h (at 37°C) under 5% CO_2, and the percentage of zygotes that cleaved to form two-cell embryos was determined. The percentage of zygotes that developed to two-cell embryos was not altered in control cultures or cultures exposed to 1.5 *M* PG (78% in both cultures), but was reduced in cultures exposed to 3.0 *M* PG (7%; $p < 0.05$). Embryonic development was inhibited completely in zygotes exposed to 6.0 *M* PG. The presence of sucrose in the incubation medium did not influence embryonic development (Damien et al., 1989). In a later publication by the same authors, the data indicated that the exposure of $B_6D_2F_1$ mouse zygotes to ≥2.5 *M* PG for 2–7 min altered both intracellular pH and developmental potential. In that these effects were independent of volume changes noted in zygotes and therefore intracellular PG concentrations, the authors postulated that the toxicity of PG is mediated by direct alteration of the cell membrane (Damien et al., 1990).

Teratogenicity

Kavlock et al. (1987) investigated the teratogenic potential of several substances, including PG. A group of 30 pregnant female CD-1 mice was given single oral doses of 10,000 ppm PG on days 8–12 of gestation. Fertility rates, numbers of maternal deaths, numbers of resorptions, average litter sizes, birth weights, and pup postnatal weight gain were monitored in an assessment of the maternal and perinatal effects of PG. The fertility rates of mice given PG were not significantly different from those of control mice. There were no maternal deaths or resorptions observed in any of the animals dosed with PG. All other parameters measured were not significantly different from control values. PG was not a teratogen in this test.

PG (dose 0.01 ml/g body wt) also was injected subcutaneously into each of 21 pregnant ICR/Jcl female mice (9–12 weeks old) on day 9, 10, or 11 of gestation. Days 9–11 correspond to the sensitive stage for the induction of fetal deaths and malformations in this strain of mice. The following malformations were noted in 5 of the 226 living fetuses: open eyelid (3 fetuses), polydactyly (1 fetus), and cleft palate (1 fetus). In a control group of 28 mice (same strain and weight range) injected subcutaneously with water (0.01 ml/g body wt) during pregnancy, the only malformation noted was exencephalus in 1 of 320 living fetuses. The incidence of malformations in 1,026 living fetuses from an untreated control group of 90 pregnant mice (same strain and weight range) was 3 fetuses with polydactyly, exencephalus, and open eyelid, respectively (Nomura, 1977).

Ascitic mouse ovarian tumor cells were used in an in vitro teratogenicity assay by Braun et al. (1982). Tumor cells were labeled with [^{3}H]thymidine in situ via an intraperitoneal injection of 0.2 mCi of radioactivity. Cells were harvested and suspended in phosphate-buffered saline (10^7 cells/ml). Various concentrations of PG were added to aliquots of the cells (all concentrations not stated) and incubated at 37°C for 30 min. The teratogenicity of the substances tested was assayed by the ability of the test substances to inhibit attachment of the tumor cells to Concanavalin A-coated plastic. The extent of attachment of the cells was measured by counting the radioactivity on the Concanavalin A-coated plastic. PG did not inhibit attachment of the tumor cells; the largest dose tested was 27,000 mg/L PG. PG was a nonteratogen in this test.

Mutagenicity

In Vitro Tests

Clark et al. (1979) assayed the mutagenicity of heat transfer fluids, including PG, according to the methods of Ames et al. (1975). PG was diluted with dimethylsulfoxide (DMSO) to final concentrations of 1–10,000 μg/plate (actual concentrations tested not stated). PG was added to tester strains TA 1535, TA 1537, TA 1538, TA 98, and TA 100 both with and without metabolic activation. PG was not mutagenic in any of the strains at any concentration tested.

PG in DMSO was tested in a reverse mutation assay (Ames et al., 1975) involving *Salmonella typhimurium* strains TA 92, TA 1535, TA 100, TA 1537, TA 94, and TA 98. The dose tested was 10 mg/plate, largest noncytotoxic dose, and cul-

tures were incubated in the presence of metabolic activation. Results were considered positive if the number of revertant colonies in experimental cultures was twice that noted in the solvent control culture. PG was not mutagenic in this assay (Ishidate et al., 1984).

Sasaki et al. (1980) investigated the mutagenic effects of 60 chemicals, including PG, using an HE 2144 human fibroblast cell line and a Don-6 Chinese hamster cell line. Concentrations of 3.805, 7.61, or 22.83 mg/ml PG in HBSS, added to 1.0 μg 5-bromodeoxyuridine (BrdU), were incubated for two cell cycles with either the human or the Chinese hamster cell line. Chromosome slides were stained with acridine orange fluorescent stain, and the frequencies of sister chromatid exchanges (SCEs) were scored. The authors reported a dose-dependent increase in the frequencies of SCEs in the Chinese hamster cell line. PG was a weak, but potential, SCE inducer in this investigation.

The mutagenicity of PG was also evaluated in a chromosomal aberrations test using a Chinese hamster fibroblast cell line. The cells were exposed to 32.0 mg/ml PG in physiological saline (maximum dose) for 48 h without metabolic activation. The maximum dose was defined as the dose at which the maximum effect was obtained. Untreated cells and solvent-treated cells served as controls; the incidence of aberrations was usually <3.0% in these cells. The incidence of polyploid cells and cells with structural chromosomal aberrations, such as chromatid or chromosome gaps, breaks, exchanges, ring formations, fragmentations, and others, were recorded for each culture. The results were considered positive if the total incidence of cells with aberrations (including gaps) was ≥10%, equivocal if the incidence was between 5 and 9.9% and negative if the incidence was ≤4.9%. Positive results were reported for PG in this test (Ishidate et al., 1984).

Abe and Sasaki (1982) investigated the mutagenicity of PG in vitro in an SCE assay. PG was tested in both human cultured fibroblasts and a cultured Chinese hamster cell line, with and without the addition of liver S9 mix as a metabolic activator. SCE was measured after the addition of BrdU. PG was not mutagenic in this assay system.

PG was tested in an in vitro DNA damage alkaline elution assay according to a modification of the procedure by Kohn et al. (1974). The procedure involved incubation of Chinese hamster lung fibroblast V79 cells, in the presence and absence of metabolic activation, with 10 m*M* PG for 1, 2, and 4 h. Prior to PG exposure, DNA in the cells was labeled with [^{14}C]thymidine. Single-stranded DNA was eluted using a single polyvinyl filter, and the recovery of radioactive DNA applied to the filter was >95%. DNA damage was not observed in PG-exposed cultures (Swenberg et al., 1976).

PG was not mutagenic in other short-term in vitro tests: chromosomal aberrations (in anaphase) in human embryonic lung cells (WI-38), mitotic recombination in *Saccharomyces cerevisiae* strain D4, and basepair substitution in *Salmonella typhimurium* strains G-46 and TA-1530 (Green, 1977).

In Vivo Test

The mutagenicity of PG was evaluated in the micronucleus test using 25 (five groups of 5) 8-week-old ddY mice. Single intraperitoneal injections of PG were

administered to four groups in doses of 2,500, 5,000, 10,000, and 15,000 mg/kg in saline, respectively. The control group was dosed with saline. Femoral marrow cells were smeared on glass slides, 1,000 polychromatic erythrocytes per mouse were scored, and the number of micronucleated polychromatic erythrocytes was recorded. Results were classified as positive only when a statistically significant difference was noted between one or more treatment group(s) and the spontaneous incidence of micronucleated polychromatic erythrocytes, and the Cochran–Armitage trend test (Armitage, 1955; Cochran, 1954) indicated a positive dose response. PG was not mutagenic at any of the doses tested (Hayashi et al., 1988).

Carcinogenicity

In Vitro Test

Pienta (1980) investigated the carcinogenic potential of PG in a hamster embryo cell transformation bioassay. Embryo cells were isolated from Syrian hamsters on day 13 of gestation. Isolated cells were separated into tubes containing 2.5×10^6 cells and frozen at −195°C prior to the experiment. Samples of the frozen cell aliquots were reconstituted on day 0 of the experiment and cultured for 6 days. On day 6 of the experiment 4 ml of PG at concentrations ranging from 0.125 to 8.0% (actual concentrations not stated) was added to the cells. Positive and negative controls were tested in each experiment. After 8 days, the cultures were investigated for the presence of any transformed colonies. No transformed colonies were reported in any of the PG-dosed cells.

In Vivo Tests

Gaunt et al. (1972) investigated the carcinogenicity of PG in a 2-year feeding study using CD strain rats (methods of study previously described in Chronic Oral Toxicity). The types and numbers of neoplasms found were documented during the study. In female rats, the largest numbers of neoplasms found were mammary gland fibroadenomas. The authors indicated that these neoplasms occurred frequently in rats of the CD strain. There were no significant differences in the incidence of neoplasms found in the PG-treated animals as compared with untreated control animals. There was no observed dose–response relationship between animals fed low and high concentrations of PG. The authors reported that no carcinogenic potential was found when concentrations up to 50,000 ppm PG were administered in the diet.

Stenback and Shubik (1974) investigated the carcinogenicity of PG using Swiss mice. Three groups of animals, 50 per group, had concentrations of 10, 50, or 100% PG (diluted in acetone where necessary) applied to the shaved dorsal skin between the flanks twice a week. Applications continued for the lifetime of the animals. All lesions and neoplasms that developed were recorded weekly during the experiment; necropsy was performed on all animals. In the group that received 10% PG, 26 of 50 animals (52%) developed neoplasms; however, skin tumors were not observed. In the 50 and 100% groups, the numbers of tumor-bearing animals were 26 of 50 (52%) and 20 of 50 (40%), respectively. The distri-

bution of the more prevalent tumors in the three treatment groups was as follows: 10% PG (15 lymphomas, 7 lung adenomas, 1 liver hemangioma, and 1 thymoma), 50% PG (13 lymphomas, 13 lung adenomas, 2 liver hemangiomas, and 2 thymomas), and 100% PG (9 lymphomas, 7 lung adenomas, 4 liver hemangiomas, and 1 thymoma). In vehicle (50 mice, 100% acetone) and positive control (0.5% 7,12-Dimethylbenzanthracene, 50 mice) groups, the percentages of tumor-bearing animals were 44 and 78%, respectively. The percentage of tumor-bearing animals in the untreated control group (150 mice) was 42%. The authors reported that there was no significant carcinogenic potential when PG concentrations up to 100% were applied to the skin of Swiss mice.

PG was not found to be carcinogenic in other carcinogenicity studies, whether the method of administration was oral, cutaneous surface application, or subcutaneous injection (Fujino et al., 1965; Dewhurst et al., 1972; Wallenius and Lekholm, 1973*a,b;* du Vivier and Stoughton, 1975; Farsund, 1978).

CLINICAL ASSESSMENT OF SAFETY

Short-Term Dermal Toxicity

An aqueous cream containing 60% PG was applied daily to seven inpatients with psoriasis (one patient was diabetic) for 5 days; doses ranged from 1.5 to 6.1 g/kg/24 h (Commens, 1990). It has been calculated that a 70-kg adult would require, in normal practice, ~44 g of cream for total body coverage twice per day, resulting in an approximate dosage of 0.35 g/kg. Total-body occlusion therapy was abandoned because three of the patients were eliminated from the study due to skin irritation. Blood was collected at the beginning of application, on day 3, and at the end of the application period for measurement of the following: serum lactate, urea, electrolytes, glucose, and osmolality levels. Two of the remaining four patients had mild renal impairment, and most patients experienced desquamation either during or after the study. No significant changes in serum osmolality and lactate were noted in any of the patients. Hyperosmolality has been induced by PG in a number of clinical settings (Glasgow et al. 1983), particularly in intensive care unit patients during the administration of nitroglycerin solutions that contain PG (Bossaert and Demey, 1987; Demey and Bossaert, 1987). Additionally, hyperosmolality has resulted from the percutaneous absorption of PG in burn patients (Fligner and Jack, 1985; Kulick et al., 1980; Bekeris et al., 1979).

Skin Irritation and Sensitization

In a review article by Catanzaro and Smith (1991), it was noted that a commonly encountered problem in patch testing, particularly with PG, is reliably differentiating an irritant reaction from an allergic reaction. This is true especially if the reaction is relatively weak. Accordingly, several authors have been unable to conclude whether patch test reactions observed in subjects represented irritation or true allergic sensitization (Blondeel et al., 1978; Romaguera et al., 1981; Hannuksela and Salo, 1986; Kinnunen and Kannuksela, 1989). These studies and the

results of other human skin irritation and sensitization tests are included in this section.

Predictive Tests

Propylene Glycol. Wahlberg and Nilsson (1984) studied the irritant effects of PG in humans (number of test subjects not indicated) using laser Doppler flowmetry. PG was applied using three different methods: single open exposures to 1.0 ml PG on the ventral aspect of the thigh with and without occlusive patches for 5–15 min and repeated open exposure to 1.0 ml PG on the skin for 12 days. Blood flow at the test site was measured and used as an indication of irritation. PG, when administered under occlusive patches, caused weak erythema at the test site; the maximum reaction was measured 26 h after exposure. Single open applications and repeated open exposure to PG did not cause any irritation reactions in this test.

These authors also used a skin fold thickness test to further study the irritant effects of PG in humans (number of test subjects not mentioned). Concentrations of 100% PG were applied to the volar forearm of the test subjects for 36 consecutive days. Skin fold measurements were made daily using a Harpenden skin fold caliper. Unexposed skin sites were used as controls. Under the conditions of this test, PG did not cause any increase in skin fold thickness in any of the test subjects.

Willis et al. (1989) studied the irritant effects of PG in 48-h patch tests. Ten adult males received patches, containing 100% PG, in Finn chambers applied to the volar aspect of the forearm for 48 h. The test sites were examined for any reaction 1 h after removal of the patches. Reactions were characterized as either negative, mild, moderate, or severe. Skin biopsies were also performed, and samples obtained were examined microscopically. Reactions were not observed when the application sites were examined macroscopically. Upon microscopic examination, half of the samples contained occasional areas of slight spongiosis, accompanied by a minimal infiltrate of mononuclear cells into the epidermis. No significant irritant effects were reported in this test.

Motoyoshi et al. (1984) conducted both 48-h patch tests and 21-day repeated insult patch tests to assess the irritation potential of PG in humans. In the 48-h patch tests, patches containing 0.05 ml 100% PG were applied to the backs of 50 adult men. After patches were removed, the sites were scored for irritation and skin samples were taken for microscopic examination. Exposure to PG produced erythema and edema in a dose-dependent manner in the test subjects (number of reactions not specified).

In 21-day patch tests by these authors, 24 adult men received patches containing 0.05 ml of 1, 3, 10, or 30% PG. Patches were applied daily to the upper backs of each of the test subjects. Sites were scored for irritation and skin samples were taken for microscopic examination. Concentrations of 10 and 30% PG produced irritation reactions in an unspecified number of subjects. The authors concluded that PG caused primary irritation reactions in this test.

Ten healthy nonatopic male volunteers were patch tested with 100% PG. The test substance was applied for 48 h to the volar aspect of the forearm using 8-mm

Finn chambers, and reactions were scored 1 h after patch removal according to the following scale: 0 (negative) to 3 (erythema, edema, and vesiculation). Half of the subjects were patch tested with distilled water, negative control, under occlusive patches. Punch biopsies were obtained from each patch test site, and skin samples were processed for immunocytochemistry and electron microscopy. Concerning the immunocytochemical technique, the antibody OKT6, which specifically recognizes the CD1a antigen expressed on the surface of the Langerhans cells and indeterminate dendritic cells in the skin, was used. The Student paired *t* test was used to compare the experimental $CD1^+$ cell density and $CD1^+$ cell dendrite length with that of the control. The majority of the reactions observed were mild to moderate, and the number and dendrite length of $CD1^+$ cells within the epidermis at PG-treated sites were not significantly different from the observations at negative control sites. The Spearman rank coefficient of correlation indicated a nonlinear correlation between $CD1^+$ cell density and clinical grading ($r = 0.1157$). However, there appeared to have been a relationship between these parameters. Mild reactions generally caused a slight decrease in density, and severe reactions caused an increase in $CD1^+$ cell number. As for dendrite length, the greater the intensity of the reaction, the shorter the dendrite (Willis et al., 1990).

Willis et al. (1988) performed an experiment to determine optimum concentrations of several compounds, including PG, which would induce irritation in 75% of a test population. Two concentrations of PG, 50 and 100%, were tested using 16 and 35 healthy volunteers, respectively. Each subject received an application of PG, under occlusive patches for 48 h, on the volar area of the forearm via a Finn chamber. At the end of the exposure period, the patches were removed and the test sites were scored 1 h later. The grading system used was − (no reaction) to 4+ (intense erythema with bullous formation). In the subjects tested with 50% PG, there were no positive reactions. Among the subjects tested with 100% PG, 14 (40%) positive reactions were reported (11 ± reactions, 3 1+ reactions). PG had marginal irritant properties in this test.

The contact sensitization potential of 12% PG in a cream vehicle was tested using a panel of 204 test subjects. In the induction phase of the experiment, 10 successive 48- or 72-h patches containing 0.5 g PG were applied under occlusive patches to the upper arms of each of the test subjects. Following a 2-week nontreatment period, a 72-h occlusive challenge patch containing 0.5 g PG was applied to the test site on each of the panelists. Reactions were scored after removal of the challenge patch. There were no reactions noted in any of the 204 panelists in this experiment (Marzulli and Maibach, 1973).

A 21-day cumulative irritancy assay was conducted by Patel et al. (1985) using PG on healthy volunteers. Webril patches containing PG (concentration not stated) were applied under occlusive patches to the back of each of 25 subjects. Patches were removed and changed daily for a total of 21 days. Test sites were examined for irritation daily prior to application of the next patch. The scale used for scoring ranged from 0 (no dermatitis) to 4 (erythema, induration, vesicles, and/or bullae covering the test site). The total cumulative irritation score was 72.0 in this test.

Trancik and Maibach (1982) investigated the irritation/sensitization potential of PG in three tests. In a cumulative irritation procedure, 24-h occlusive patches containing 0.2 ml PG were applied to the backs of 10 subjects. A total of 21 consecutive patches were applied. Test sites were observed for reactions daily. In this 21-day test, one subject had an equivocal reaction on day 20. No other reactions were noted.

These authors continued their investigation of the irritation/sensitization potential of PG in a modified Draize sensitization test. In the induction phase, a panel of 203 healthy volunteers had 48-h occlusive patches containing 0.2 ml PG applied to their backs. A total of 10 applications were made over a 21-day period. After a 2-week nontreatment period, challenge patches containing 0.2 ml PG were applied to fresh test sites and removed 48–72 h later. Reactions were scored on a scale of 0.0–4.0. During the induction phase, equivocal reactions (score = 0.5) were noted in eight of the panelists. Upon challenge, 19 cutaneous reactions were noted (6 had 0.5 reactions, 6 had 1.0 reactions, and 7 had 2.0 reactions).

The same authors also conducted a provocative use test with PG using the 19 panelists with cutaneous reactions in the sensitization test described. Applications of 0.1 ml PG were made twice daily to the cubital fossa of each subject for a total of 7 days. Sites were scored for reactions on day 8. There were no reactions noted in any of the panelists. It was concluded that the cutaneous reactions observed in 19 subjects were irritant reactions.

The results of predictive clinical tests on PG are summarized in Table 11.

Polypropylene Glycol. A total of 300 human subjects received continuous and repeated dermal applications of undiluted PPG 2000. Details concerning the experimental procedure were not included. The test substance caused neither skin irritation nor sensitization (American Industrial Hygiene Association, 1980).

Provocative Tests

A total of 866 patients with various dermatological conditions were patch tested (closed or covered patches) with 100% PG from April 1951 to April 1952. The patches were applied to clinically normal skin, and test sites were examined 48 h after patch application. Positive reactions were observed in 138 (15.7%) patients. Reactions ranged from simple erythema (+) to erythema with induration and vesiculation (+ + + +). Eighty-nine of the 138 patients with positive reactions suffered from dermatitis venenata. A seasonal fluctuation in the incidence of positive reactions was also noted. The incidence was at a minimum when the climate was hot and humid (July, August, and September 1951 in New York City) and significantly greater during the cooler and less humid seasons. Of the 84 patients involved with simultaneous testing with several samples of PG from different sources, positive reactions were observed in 15. There were no differences in patient responses to different brands of PG (Warshaw and Herrmann, 1952).

Twenty-three of the 138 patients with positive reactions to 100% PG in the preceding study were patch tested with aqueous PG. There were only five positive

TABLE 11. *Clinical predictive patch testing to evaluate skin irritation and sensitization potential of Propylene Glycol (PG)*

Test substance	No. of subjects	Procedure	Results	Ref.
100% PG	Not stated	Single 5- to 15-min exposures to 1 ml of PG (open and under occlusion)	Weak erythema at test site (under occlusion); no reactions (open)	Wahlberg and Nilsson (1984)
100% PG	10	48-h patch test using Finn chamber	No significant skin irritation	Willis et al. (1989)
100% PG	50	48-h patch test; 0.05-ml application	Erythema and edema noted	Motoyoshi et al. (1984)
100% PG	10	48-h patch test using Finn chamber	Mild to moderate erythema predominated	Willis et al. (1990)
100% PG	Not stated	Repeated open exposures to 1 ml PG for 12 days	No irritation	Wahlberg and Nilsson (1984)
100% PG	Not stated	Repeated open exposures for 36 consecutive days	No increase in skinfold thickness	Wahlberg and Nilsson (1984)
PG (50 and 100%)	16 (50% PG group); 36 (100% group)	48-h exposure, under occlusive patches	No reactions to 50% PG; 11 ± and three 1+ reactions to 100% PG	Willis et al. (1988)
PG (1, 3, 10 and 30%)	24	Repeated closed patch applications for 21 days	Primary irritation with 10 and 30% PG	Motoyoshi et al. (1984)
12% PG in cream vehicle	204	10 successive 48- or 72-h occlusive patches (induction); 72-h occlusive challenge patch	No reactions	Marzulli and Maibach (1973)
PG	25	Repeated exposures, under occlusive patches, for 21 days	Total cumulative irritation score of 72	Patel et al. (1985)
PG	10	Repeated exposures, 0.2 ml under occlusive patches, for 21 days	1 subject with equivocal reaction	Trancik and Maibach (1982)
PG	203	Modified Draize sensitization test; 10 0.2-ml applications under occlusive patches (induction); 48- to 72-h challenge	13 positive reactions; 6 equivocal reactions	Trancik and Maibach (1982)
PG	19 (subjects with positive reactions in preceding study)	Provocative use test; 0.1-ml applications twice daily for 7 days	Cutaneous reactions noted in preceding study confirmed as irritant reactions	Trancik and Maibach (1982)

responses to 10% PG, and the application of 2.5% PG in water to 3 of the 23 patients resulted in one positive reaction. Additionally, 16 of the patients with positive reactions to 100% PG were also tested by simple inunction of the test substance. There was no evidence of an inflammatory response to the inunction either shortly after application or 48 h later (Warshaw and Herrmann, 1952).

When 1,556 patients were patch tested with 100% PG, positive reactions were observed in 194 subjects. Four patients had "true allergy" and the remainder had irritant reactions. Three groups of 42 patients with positive reactions to 100% PG

were later tested with 3.2, 10, and 32% PG, respectively, and the results were as follows: 3.2% PG (9 positive reactions), 10% PG (12 positive reactions), and 32% PG (20 positive reactions) (Hannuksela et al., 1975; Catanzaro and Smith, 1991). Details concerning the experimental procedure were not included in the review article by Catanzaro and Smith (1991).

The irritation/sensitization effects of PG were investigated using a panel of 38 patients with histories of PG reactions. Finn chambers containing 2, 10, 32, or 100% PG were applied to the back of each of the test subjects. Exposure lasted from 20 to 24 h, and reactions were scored, 1, 2, 4, and 5 days postexposure. Peroral challenge doses of 2% PG, either 2 or 5 ml, were administered to each of the test subjects. Reactions were recorded for 24 h or until compound-related signs had resolved. In the initiation phase of the experiment, allergic eczema was reported in 11 of 38 patients. Other types of eczema were noted in 16 of 38 patients. In 15 of the subjects who reacted to PG in the initiation phase, peroral challenge resulted in extensive exanthema, which resolved spontaneously within 48 h after challenge. The authors calculated that, based on the results of this experiment, 1% of all patients with eczema "may suffer harmful reactions either from the internal or external use of PG" (Hannuksela and Forstrom, 1978).

In another study, positive reactions were observed in 12 of 84 patients patch tested with 100% PG. Five subjects had allergic reactions, and seven had skin irritation reactions (Andersen and Storrs, 1982).

Nater et al. (1977) investigated the irritant effects of PG in 48-h patch tests. A group of 98 subjects, all with histories of allergic contact dermatitis, were patch tested with 100% PG. Patches containing an unspecified volume of PG were applied to the skin on the backs of each of the test subjects. After the patches were removed, sites were scored for reactions (scale = 0 to 3+), and skin samples were obtained by biopsy for microscopic examination. There were 11 1+ reactions noted out of the 98 subjects tested. Microscopic examination of skin samples from subjects with positive reactions indicated edema and mononuclear perivascular infiltrate in eight of the samples. The authors concluded that the reactions observed were primary irritation reactions.

The irritation/sensitization potential of PG or hexylene glycol was investigated by Kinnunen and Hannuksela (1989) using several different methods. Occlusive patches containing 30% PG were applied (48 h) to the skin of 823 patients with histories of contact dermatitis. Skin reactions were scored 1–4 h and 2 days after removal of the patches. Edema and erythema were noted in 3.8% of the patients tested. Erythema alone was noted in 7.4% of the tested patients.

Additional patch tests were conducted by these authors on 22 patients who had edema/erythema reactions to PG or hexylene glycol. Serial dilutions of 1, 2, 10, and 30% PG were applied using the same methods described. A positive reaction was reported in one patient tested with 1% PG; three patients tested with 30% PG had positive reactions.

The effect of PG on transepidermal water loss (TEWL), skin blood flow, and nonimmunologic contact reactions (NICR) to benzoic acid was also tested by Kinnunen and Hannuksela using a spearate panel of 8 dermatitis patients and 11 normal volunteers. Doses of 20 μl 50% PG were applied to the backs of the 19

panelists. The eight patients received five applications at the same time; the normal volunteers received repeated applications over a 7-day period. TEWL was measured with an evaporimeter after the first applications to patients and after the last applications to normal volunteers. After a 20-μl application of benzoic acid to the test site, the NICR and blood flow were measured. PG significantly increased TEWL on day 3 in the patients with dermatitis and on day 8 in the normal volunteers. Cutaneous blood flow was increased in the test subjects. The NICRs in the test subjects were not significantly different from controls. PG had irritant properties in these tests.

A total of 183 patients were patch tested with 38% PG. Allergic reactions were observed in 23 patients (Huriez et al., 1966; Catanzaro and Smith, 1991). Details concerning the experimental procedure were not included in the review article by Cantanzaro and Smith (1991).

A total of 86 contact dermatitis patients were tested (patch tests and repeated open application tests) with PG as well as with other common ingredients of dermatological preparations. In initial patch tests, 30.0% PG in water was applied for 24 h, under occlusive patches, using Finn chambers. Reactions were scored 0.3–2 h after patch removal according to the International Contact Dermatitis Research Group grading scale (Wilkinson et al., 1970): ?+ (doubtful reaction) to +++ (extreme reaction). In this study, + (weak, nonvesicular) and ++ (strong, edematous or vesicular) reactions were classified as positive. Nineteen patients with allergic reactions to 30% PG in initial patch tests were subsequently patch tested (same procedure) as follows: 1% aqueous PG (5 patients), 10% aqueous PG (2 patients), and 30% aqueous PG (12 subjects); all patients had + or ++ reactions. Repeated open application tests (ROATs) were also performed on these patients. In these tests, 5% PG in a cream base was applied to a 5 × 5–cm cream on the flexor aspect of the forearm twice daily for 7 days; the cream base contained vegetable oil, cetylstearyl alcohol, parabens, and 85% water. Positive reactions to 5% PG were observed in all 5 patients who had positive reactions to 1% PG, 1 of 2 patients with positive reactions to 10% PG, and 4 of 12 patients with positive reactions to 30% PG (Hannuksela and Salo, 1986).

Three of 78 patients patch tested with 10% PG had allergic reactions (Braun, 1969; Cantanzaro and Smith, 1991). In other studies, allergic reactions were observed in 2 of 100 patients (Fisher et al., 1971; Cantanzaro and Smith, 1991) and 13 of 330 patients (Blondeel et al., 1978; Cantanzaro and Smith, 1991) patch tested with 10% PG. Details concerning the experimental procedures for these tests were not included in the review article by Cantanzaro and Smith (1991).

In a study by the North American Contact Dermatitis Group (Adams et al., 1985), 29 cutaneous reactions were observed in a population of 399 patients with cosmetic-related contact dermatitis that was patch tested (Finn chambers) with 10% aqueous PG.

At test concentrations of 5 and 10% PG, 15 of 1,450 patients had positive patch test reactions (Romaguera et al., 1981). Details concerning the experimental procedure were not stated.

In a series of patch tests from 1968 to 1983, Angelini et al. (1985) reported a total of 27 positive reactions in 3,364 patients tested with 5% aqueous PG.

Test reactions to standard patch test allergens were evaluated using 1,040 subjects (851 atopic patients and 189 healthy age-matched controls). The atopic patients (Group I, 28–41 years old; Group II, 19–27 years old) were classified in a total of eight groups as follows: Group I 1 (79 patients) and Group II 1 (97 patients), patients with severe atopic dermatitis who had been hospitalized; Group I 2 (149) and Group II 2 (213), patients who had been treated as outpatients; Group I 3 (59) and Group II 3 (74), patients with mild atopic dermatitis and allergic rhinitis, allergic conjunctivitis, or asthma; and Group I 4 (110) and Group II 4 (70), patients who had been examined at the hospital for allergic rhinitis, allergic conjunctivitis, or asthma without dermatitis. PG (5%) was applied, using a Finn chamber, to the upper back of each patient for 2 days, and reactions were scored on day 3. Each test site was free of dermatitis at the time of patch testing and dermatitis at other sites was said to have been in an inactive phase. Of the 851 patients patch tested with 5% PG, 2.5% had allergic reactions and 3.9% had irritant/follicular reactions (Lammintausta et al., 1992).

In an epicutaneous chamber test, Hannuksela et al. (1976) investigated the irritant effects of PG. Concentrations of 2.0% PG in water were placed in chambers on the back of each of 880 test subjects. The exposure time lasted from 20 to 24 h. After removal of the test chambers, the exposed sites were scored at 30 min, 2, 4, and 5 days. At the concentration tested, two (0.2%) positive reactions were noted among the panelists. The authors noted that PG often caused irritant reactions when tested under occlusive patches and that the methods employed in this test indicated a true value for allergy to the compound.

The effects of PG and other compounds used as vehicles were investigated by Mendelow et al. (1985). PG increased the allergic responses in 43 patients patch tested with 50 μg of 1% nickel sulfate solution. The increase in reactions noted was greater than the number of patients who had an allergic reaction to 50% PG alone. The data indicated that PG, when used as a vehicle, influenced the amount of nickel sulfate exposed to the skin, having modified the allergic reaction potential. In the order of largest to smallest induction of allergic responses, the vehicles were ranked: DMSO > PG > cetomacrogol cream > yellow soft paraffin.

Several additional studies concerning allergy to PG have been published (Broeckx et al., 1987; Agren-Jonsson and Magnussen, 1976; Fastner, 1981; Fisher and Branaccio, 1979; Angelini and Meneghini, 1981; Oleffe et al., 1979; Wahlberg, 1984).

The results of clinical provocative tests on PG are summarized in Table 12.

Case Reports

A 26-year-old woman applied minoxidil to an area of diffuse alopecia located immediately above the nape of her neck. After 6 weeks of application, a pruritic papular eruption was noted in the treated area and nape of the neck. Patch test reactions to the minoxidil solution and 5% aqueous PG were positive. There was no reaction to a 2% minoxidil solution in which glycerin was the solvent (Fisher, 1990).

A pruritic, macular, and papular eruption was observed on the scalp (vertex

TABLE 12. *Clinical provocative patch testing to evaluate skin irritation and sensitization potential of Propylene Glycol (PG)*

Test substance	No. of patients	Procedure	Results	Ref.
PG (2.5, 10, and 100%)	866 dermatologic patients	48-h patch test (closed or covered patches)	Skin irritant (138 positive reactions); 5/23 and 1/3 positive patients had positive reactions to 10 and 2.5% PG	Warshaw and Herrmann (1952)
PG (3.2, 10, 32, and 100%)	1,556 patients	Patch test	True allergy (4 patients) and irritation (1,552 patients); results for 3 groups of 42 positive patients patch tested with PG: 3.2% PG [9 positive (+) reactions], 10% PG (12+), and 32% PG (20+)	Hannuksela et al. (1975); Catanzaro and Smith (1991)
PG (2, 10, 32, and 100%)	38 patients	Simultaneous 20- to 24-h Finn chamber applications; per-oral challenge dose of 2% PG	Allergic eczema and other types of eczema in 11 and 16 patients during initiation; challenge of 15 patients selected caused extensive exanthema that cleared within 48 h	Hannuksela and Forstrom (1978)
100% PG	84 patients	Patch test	Allergic reactions (5 patients) and skin irritation (7 patients)	Andersen and Storrs (1982)
100% PG	98 eczema patients	48-h patch test	Primary irrtation; 11 1+ reactions	Nater et al. (1977)
50% PG	8 patients with allergic contact dermatitis; 11 normal subjects	5 simultaneous 20-μl applications per patient; repeated applications over 7-day period in normal subjects; at end of exposure, benzoic acid applied to evaluate nonimmunologic contact reactions (NICRs) to benzoic acid	PG had irritant properties; it significantly increased transepidermal water loss and increased cutaneous blood flow in both groups; NICRs were not significantly different from controls	Kinnunen and Hannuksela (1989)
38% PG	183 patients	Patch test	23 allergic reactions	Huriez et al. (1966); Catanzaro and Smith (1991)

PG (30% aqueous)	86 contact dermatitis patients	24-h occlusive patch applications (Finn chambers) for 7 days	Allergic reactions in 19 patients	Hannuksela and Salo (1986)
PG (1, 10, and 30% in water; 5% in cream)	19 patients with allergic reactions (from preceding study)	24-h occlusive patch applications of 1, 10, and 30% PG (Finn chambers) for 7 days; for 5% PG, repeated open application test (0.1-ml applications twice daily for 7 days)	Positive reactions: 1% PG (5 subjects with + or + + reactions); 10% PG (2 with + or + + +), 30% PG (12 with + or + + +); incidence of + reactions to 5% PG in preceding groups: ¾ (1% PG group); ½ (10% PG group); and 4/12 (30% PG group)	Hannuksela and Salo (1986)
30% PG	823 patients	48-h occlusive patch application	Erythema in 7.4% of patients; erythema and edema in 3.8% of patients	Kinnunen and Hannuksela (1989)
PG (1, 2, 10, and 30%)	22 patients (22 of patients with erythema and edema in preceding study)	48-h occlusive patch application	Positive reactions: 1% PG (1 patient), 30% PG (3 patients)	Kinnunen and Hannuksela (1989)
10% PG	78 patients	Patch test	3 allergic reactions	Braun (1969); Cantanzaro and Smith (1991)
10% PG	100 patients	Patch test	2 allergic reactions	Fisher et al. (1971); Cantanzaro and Smith (1991)
10% aqueous PG	399 patients	48-h patch test using Finn chambers	29 cutaneous reactions	Adams et al. (1985)
10% PG	330 patients	Patch test	13 allergic reactions	Blondeel et al. (1978); Cantanzaro and Smith (1991)
5 and 10% PG	1,450 patients	Patch test	15 positive reactions	Romaguera et al. (1981)
5% aqueous PG	3,364 patients	Patch tests from 1968 to 1983	27 positive reactions	Angelini et al. (1985)
5% aqueous PG	851 atopic patients	Finn chamber application for 2 days	2.5% of patients had allergic reactions; 3.9% had irritant/follicular reactions	Lammintausta et al. (1992)
2% aqueous PG	880 patients	Epicutaneous chamber test; chambers applied for 20–24 h	2 allergic reactions	Hannuksela et al. (1976)

region) of a man who had applied minoxidil to this area for 1 month. Patch test reactions to the minoxidil solution and 5% PG were positive. However, there was no reaction to 5% minoxidil in glycerin. The eruption recurred when 5% PG was rubbed into a small area of the scalp after the initial reaction subsided (Fisher, 1990).

A 52-year-old woman suffered from chronic otitis media for 2 years and had used various ear drops and ointments. During the last 2 weeks of the 2-year episode, the patient used eardrops containing 2.5% hydrocortisone in a mixture of PG (50%) and water. Patch test results indicated a papulovesicular reaction (+ + +) to this product after 72 h. There were no reactions to 10% aqueous PG and 1 and 10% hydrocortisone in ethanol. However, oral challenge with 5 ml of PG caused a pruritic macular rash on the abdomen, a flare-up of the external ear dermatitis, and a flare-up of the patch test reaction on the back within 20 h. When the patient was patch tested 1 week after the oral challenge, a + reaction to 50% PG and a + + reaction to undiluted PG were noted after 72 h (Frosch et al., 1990).

Several case studies have been published, documenting assorted reactions to PG administration in humans (Arulanantham and Genel, 1978; Demey et al., 1988; Fligner et al., 1985; Kelner and Bailey, 1985; Lolin et al., 1988; MacDonald et al., 1987; Martin and Finberg, 1970).

SUMMARY

PG is an aliphatic alcohol and PPG is a polymer of PG and water. In cosmetics, PG is used as a skin-conditioning agent–humectant, solvent, viscosity-decreasing agent, and humectant. PPGs are used as miscellaneous skin-conditioning agents. Product formulation data submitted to FDA in 1984 indicated that PG was used in a total of 5,676 cosmetic products at concentrations up to >50%. Data (FDA, 1984) submitted on PPGs were as follows: PPG 9 (6 products), PPG 26 (10 products), and PPG 425 (1 product). All three ingredients were used at concentrations up to 5%.

While concentration of use data are no longer reported to FDA by the cosmetics industry, current frequency of use data reported indicate that PG is used in a total of 4,892 cosmetic products and PPG 9 in 5 products. There are no reported uses of PPG 26 and PPG 425. Current data on PG supplied to the CTFA indicate that concentrations of use range between 3 and 5% in products manufactured by one company.

In mammals, the major route of PG metabolism is to lactaldehyde and then lactate via hepatic alcohol and aldehyde dehydrogenases. When PG was administered intravenously to human subjects (patients), elimination from the body occurred in a dose-related manner. The results of animal studies on PPGs 425, 1025, and 2025 indicate that they are readily absorbed from the GI tract and are excreted in the urine and feces.

The cytotoxicity of human natural killer cells was decreased significantly in an assay in which target cells (cultured K562 erythroleukemia cells) were incubated with 1% PG.

PG was relatively harmless (LD_{50} = 21 g/kg) in acute oral toxicity studies

involving rats. Acute oral toxicity studies on PPGs of various molecular weights (300–3,900) have indicated LD_{50} values (rats) ranging from 0.5 to >40 g/kg.

In acute dermal toxicity studies involving groups of five albino rabbits, doses of PPG 1025 (20 ml/kg) and PPG 2025 (20 ml/kg) did not cause death. Two of five rabbits dosed with 20 ml/kg PPG 425 and one of five dosed with 10 ml/kg PPG 425 died.

In subchronic oral toxicity studies, PPG 2000 induced, at most, slight decreases in growth and body weight effects in rats. PPG 750 caused slight increases in liver and kidney weights in rats. Following the subchronic oral administration of PPG 750 to dogs, slight increases in liver and kidney weights were noted.

In a subchronic dermal toxicity study (rabbits), PPG 2000 did not cause any adverse effects at doses of 1 ml/kg. Slight depression of growth was observed after the administration of 5- and 10-ml/kg doses.

Test substance–related lesions were not observed in rats that were fed diets containing 50,000 ppm PG (2.5 g/kg/day) for 15 weeks or in rats that were fed PG concentrations up to 50,000 ppm in the diet for 2 years. Similar results were reported in a study in which dogs were fed 2 or 5 g/kg PG in the diet for ~103 weeks. In another subchronic study, dogs received 5% PG in drinking water for 5–9 months. The results of tests for hepatic and renal impairment were negative. However, in cats fed diets containing PG, erythrocyte destruction was noted at concentrations as low as 6% PG.

PG did not induce corneal damage in rabbits in the Draize test and was classified as a slight ocular irritant in another ocular irritation study. PPGs 425, 1025, and 2025 were classified as harmless agents in rabbits in another ocular irritation study; PPG 1200 induced slight, transient ocular irritation in an albino rabbit.

In a 24-h skin irritation test involving nude mice, there were no reactions to 10% PG. Hypertrophy, dermal inflammation, and proliferation were observed at a concentration of 50% PG.

Draize test results indicated that PG was, at most, a mild skin irritant when applied for 24 h to abraded and intact skin of rabbits. When PG was applied to the skin of guinea pigs and rabbits (guinea pigs and rabbits lack sweat glands) for 48 h using open and closed patches, no reactions were observed. The results of 48-h and 21-day open and closed patch tests involving Gottingen swine (no sweat glands) indicated no reactions to PG.

Results were negative for 100% PG in a mouse external ear swelling sensitization test. The results of a GPMT, OET, and chamber (Finn chamber) test indicated no sensitization reactions to 70% PG. In another maximization test, PG was classified as a potentially weak sensitizer. The results of six other guinea pig sensitization tests indicated that PG was not an allergen.

Single and repeated applications of PPG 425, PPG 1025, and PPG 2025 did not cause skin irritation in the rabbit. Repeated applications of PPG 1200 to rabbits caused mild reactions at abraded skin sites and no reactions at intact sites.

PG was not teratogenic in female CD-1 mice when administered at a concentration of 10,000 ppm on days 8–12 of gestation. Malformations were observed in 5 of 226 living fetuses from female mice injected subcutaneously with PG (dose = 0.1 mg/g body wt on day 9, 10, 11 of gestation). However, 3 fetuses with malfor-

mations were also noted among 1,026 living fetuses from the untreated control group of pregnant mice.

In a continuous breeding reproduction study, there were no significant differences between control and experimental groups of albino mice with respect to the following: mating index, fertility index, mean number of live pups per litter, proportion of pups born alive, and sex of pups born alive.

Embryonic development was reduced in cultures of mouse zygotes exposed to 3.0 *M* PG and inhibited completely in cultures exposed to 6.0 *M* PG for 20 min.

In the Ames test, PG was not mutagenic in strains TA 1535, TA 1537, TA 1538, TA 98, and TA 100 of *Salmonella typhimurium* with and without metabolic activation. PG caused a dose-dependent increase in the frequency of SCEs in a Chinese hamster cell line and was classified as a weak inducer of SCEs. In another study, PG was not mutagenic when tested in an SCE assay involving human cultured fibroblasts and a cultured Chinese hamster cell line both with and without metabolic activation. Chromosomal aberrations were induced in Chinese hamster fibroblasts in another assay. PG was not mutagenic in additional in vitro tests: chromosomal aberrations, mitotic recombination, basepair substitution, micronucleus test, reverse mutation, and DNA damage.

PG disturbed the proliferation of urinary bladder epithelial cells from the rat, having reduced DNA production in tetraploid cells 1 and 2 months after the rats were injected subcutaneously. This effect was not observed at 3 months.

The results were negative when PG was tested in the hamster embryo cell transformation bioassay. In a 2-year feeding study involving CD strain rats, PG was not carcinogenic when concentrations up to 50,000 ppm were administered in the diet. In a lifetime dermal carcinogenicity study, three groups of Swiss mice received dermal applications of 10, 50, and 100% PG, respectively. The tumor incidence in each of the three groups did not differ from that noted in the negative control group; skin tumors were not observed.

PG induced skin irritation and sensitization reactions in normal subjects and in patients. In these studies test concentrations ranged from 2 to 100% PG. Reactions were observed at concentrations as low as 10% PG in predictive tests and as low as 2% in provocative tests.

PG also increased the allergic responses in 43 patients patch tested with 50 μg of 1% nickel sulfate solution. Neither skin irritation nor sensitization reactions were observed in 300 subjects who received continuous and repeated dermal applications of undiluted PPG 2000.

DISCUSSION

Because of the results of human irritation and sensitization tests, establishing a concentration limit for PG is considered necessary. Both provocative and predictive test data were considered in the process of making the final determination.

In provocative tests, allergic reactions were observed in 2 of 880 (0.2%) eczema patients patch tested with 2% aqueous PG, in 13 of 330 (4%) patients patch tested with 10% PG, and in 21 of 851 (2.5%) atopic patients patch tested with 5% PG. Thirty-three (3.9%) of the 851 atopic patients also had irritant/follicular reactions.

Furthermore, in a study of the North American Contact Dermatitis Group, 29 cutaneous reactions were observed in a population of 399 patients with cosmetic-related dermatitis who were patch tested with 10% aqueous PG. Patients with diseased skin may be at risk with respect to developing irritation/sensitization reactions to PG, even at low concentrations.

Predictive tests are considered more appropriate in establishing concentration limits that are based on skin irritation or sensitization data. In these tests, normal subjects were patch tested with PG concentrations ranging from 1 to 100%. In one of the studies, skin irritation was observed when 100% PG was tested under occlusive patches, but was not observed in open patch tests. When 50 and 100% PG were tested under occlusive patches on 16 subjects, there were no reactions to 50% PG and three 1+ reactions to 100% PG. In other studies, 24 normal subjects patch tested (closed patches) with 1, 3, 10, and 30% PG had skin irritation reactions only at concentrations of 10 and 30%, and neither skin irritation nor sensitization was observed in 204 subjects patch tested with 12% PG (under occlusive patches) in a cream vehicle. These studies indicate that in normal subjects, PG may be a skin irritant when tested under occlusive patches and that the skin irritation potential of this ingredient may be concentration dependent.

CONCLUSION

On the basis of the data included in this report, the CIR Expert Panel concludes that Propylene Glycol and Polypropylene Glycols are safe for use in cosmetic products at concentrations up to 50.0%.

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REFERENCES

Abe S, Sasaki M. (1982) SCE as an index of mutagenesis and/or carcinogenesis. *Prog Top Cytogenet* 2:461–514.

Adams RM, Maibach HI, Clendenning WE, et al. (1985) A five-year study of cosmetic reactions. *J Am Acad Dermatol* 13:1062–9.

Agren-Jonsson S, Magnussen B. (1976) Sensitization to propantheline bromide, trichlorocarbanilide, and propylene glycol in an antiperspirant. *Contact Dermatitis* 2:79–80.

Ahluwalia P, Amma KP. (1984) Hypolipidemic effect of propane-1,2-diol on the morphology of rat erythrocytes. *Res Bull Panjab Univ Sci* 35:157–60.

AMA Division of Drugs. (1983) *AMA Drug Evaluations*. 5th ed. Chicago: American Medical Association, 1391.

American Industrial Hygiene Association. (1980) Workplace environmental exposure level guide. Polypropylene glycols. *Am Ind Hyg Assoc J* 41:A53–5.

Ames BN, McCann J, Yamasaki E. (1975) Methods for detecting carcinogens and mutagens with the salmonella/mammalian-microsome mutagenicity test. *Mutat Res* 31:347–64.

Amma KP, Namagiri T, Chum A. (1984) The redox state of propane-1,2-diol fed rat erythrocytes. *Res Bull Panjab Univ Sci* 35:109–14.

Andersen, KE, Storrs FJ. (1982) Hautreizungen durch propyleneglykol. *Hautarzt* 33:12–4.

Angelini G, Meneghini CL. (1981) Contact allergy from propylene glycol. *Contact Dermatitis* 7:197–8.

Angelini G, Vena GA, Meneghini CL. (1985) Allergic contact dermatitis to some medicaments. *Contact Dermatitis* 12:263–9.

Armitage P. (1955) Test for linear trends in proportions and frequencies. *Biometrics* 11:375.

Arulanantham K, Genel M. (1978) Central nervous system toxicity associated with ingestion of propylene glycol. *J Pediatr* 93:515–6.

Bartsch W, Sponer G, Dietman K, Fuchs G. (1976) Acute toxicity of various solvents in the mouse and rat. LD_{50} of ethanol, diethylacetamide, dimethylformamide, dimethyl-sulfoxide, glycerine, *N*-methylpyrrolidone, polyethylene glycol 400, 1,2-propanediol and Tween 20. *Arzneimittelforsch* 26:1581–3.

Bauer MC, Weiss DJ, Perman V. (1992) Hematologic alterations in adult cats fed 6 or 12% propylene glycol. *Am J Vet Res* 53:69–72.

Bekeris L, Baker C, Fenton J, et al. (1979) Propylene glycol as a cause of elevated serum osmolality. *Am J Clin Pathol* 72:633–6.

Blondeel A, Oleffe J, Achten G. (1978) Contact allergy in 330 dermatological patients. *Contact Dermatitis* 4:270–6.

Bossaert L, Demey H. (1987) Propylene glycol intoxication. *Arch Intern Med* 147:611–2.

Braun W (1969) Kontaktallergien gegen poylaethylenglykole. *Z Haut Geschlechstkr* 44:385–8.

Braun AG, Buckner CA, Emerson DJ, Nichinson BB. (1982) Quantitative correspondence between the in vivo and in vitro activity of teratogenic agents. *Proc Natl Acad Sci USA* 79:2056–60.

Brazeau GA, Fung H-L. (1989) Use of an in vitro model for the assessment of muscle damage from intramuscular injections: in vitro—in vivo correlation and predictability with mixed solvent systems. *Pharm Res* 6:766–71.

Broeckx W, Blondeel A, Dooms-Goossens A, Achten G. (1987) Cosmetic intolerance. *Contact Dermatitis* 16:189–94.

Budden R, Kuehl UG, Bahlsen J. (1979) Experiments on the toxic, sedative, and muscle relaxant potency of various drug solvents in mice. *Methods Toxicol* 102:467–74.

Catanzaro JM, Smith JG. (1991) Propylene glycol dermatitis. *J Am Acad Dermatol* 24:90–5.

Christopher MM, Perman V, Eaton JW (1989) Contribution of propylene glycol induced Heinz body formation to anemia in cats. *J Am Vet Med Assoc* 194:1045–56.

Christopher MM, Eckfeldt JH, Eaton JW. (1990*a*) Propylene glycol ingestion causes D-lactic acidosis. *Lab Invest* 62:114–18.

Christopher MM, White JG, Eaton JW. (1990*b*) Erythrocyte pathology and mechanisms of Heinz body-mediated hemolysis in cats. *Vet Pathol* 27:299–310.

Clark CR, Marshall TC, Merickel BS, Sanchez A, Brownstein DG. (1979) Toxicological assessment of heat transfer fluids proposed for use in solar energy applications. *Toxicol Appl Pharmacol* 51: 529–35.

Cochran WG. (1954) Some method for strengthening the common χ^2 test. *Biometrics* 10:417.

Code of Federal Regulations. (April 1, 1984) Title 21, Part 720.4. Voluntary filing of cosmetic product ingredient and cosmetic raw material composition statement. Information requested about cosmetic products. Washington, D.C.: U.S. Government Printing Office.

Code of Federal Regulations. (April 1, 1984) Title 21, Parts 169.175, 582.1666. Washington, D.C.: U.S. Government Printing Office.

Code of Federal Regulations. (April 1, 1984) Title 21, Part 175.300. Indirect food additives: adhesives coating and components. Resinous and polymeric coatings. Washington, D.C.: U.S. Government Printing Office.

Commens CA. (1990) Topical propylene glycol and hyperosmolality. *Br J Dermatol* 122:77–80.

Cosmetic, Toiletry, and Fragrance Association. (1992) Submission of unpublished data by CTFA. Concentration of use data on Propylene Glycol.*

Damien M, Luciano AA, Peluso JJ. (1989) Propanediol-induced alterations in membrane integrity, metabolism and developmental potential of mouse zygotes. *Hum Reprod* 4:969–74.

Damien M, Luciano AA, Peluso JJ. (1990) Propanediol alters intracellular pH and developmental potential of mouse zygotes independently of volume change. *Hum Reprod* 5:212–6.

Dean ME, Stock BH. (1974) Propylene glycol as a drug solvent in the study of hepatic microsomal enzyme metabolism in the rat. *Toxicol Appl Pharmacol* 28:44–52.

Demey H, Bossaert L. (1987) Propylene glycol intoxication and nitroglycerin therapy. *Crit Care Med* 15:540.

Demey HE, Daelmans RA, Verpooten GA, De Broe ME. (1988) Propylene glycol-induced side effects during intravenous nitroglycerin therapy. *Intensive Care Med* 14:221–6.

Denning DW, Webster AD. (1987) Detrimental effect of propylene glycol on natural killer cell and neutrophil function. *J Pharm Pharmacol* 39:236–8.

Descotes J. (1988) Identification of contact allergens: the mouse ear sensitization assay. *J Toxicol Cutaneous Ocul Toxicol* 7:263–72.

Dewhurst F, Kitchen DA, Calcutt G. (1972) The carcinogenicity of some 6-substituted benzo(a)pyrene derivatives in mice. *Br J Cancer* 26:506–8.

Dorman DC, Haschek WM. (1991) Fatal propylene glycol toxicosis in a horse. *J Am Vet Med Assoc* 198:1643–4.

Dossou G, Sicard C, Kalopissis G, Raymond D, Schaefer H. (1985) Method for assessment of experimental allergy in guinea pigs adapted to cosmetic ingredients. *Contact Dermatitis* 13:226–34.

Douglas WH, Spilman SD. (1983) In vitro ocular irritancy testing. *Altern Methods Toxicol* 1:205–30.

Dow Chemical Co. (1975) Sales specifications: U.S.P. and industrial Propylene Glycol. Midland, MI.

Dow Chemical Co. (1978) Material safety data sheet: Propylene Glycol U.S.P. and Propylene Glycol industrial. Midland, MI.

Dow Chemical Co. (1992) Submission of unpublished data by CTFA. UV spectral analysis of propylene glycol. (4 pages).*

Dow U.S.A. (May 12, 1993) Letter to Dr. Robert Elder. Comment on propylene glycol tentative final report.

Draize JH, Woodard G, Calvery HO. (1944) Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes. *J Pharmacol Exp Ther* 82:377–90.

du Vivier A, Stoughton RB. (1975) An animal model for screening topical and systemic drugs for potential use in the treatment of psoriasis. *J Invest Dermatol* 65:235–7.

Eastman Chemical Co. (1992) Submission of unpublished data by CTFA. UV spectral analysis of propylene glycol. (1 page).*

EEC Cosmetics Directive. (1990) Proposal for a Council Directive on the approximation of the laws of the member states relating to cosmetic products. *Official J Eur Communities* 21:322–77.

Eichbaum FW, Yasaka WJ. (1976) Antiarrhythmic effect of solvents: propylene glycol, benzyl alcohol. *Basic Res Cardiol* 71:355–70.

Emmanuel B. (1976) Effects of 1,3-butanediol and 1,2-propane-diol on growth, blood, metabolites, and liver glycogen in broiler chickens. *Poult Sci* 55:2289–94.

Estrin NF, Crosley PA, Haynes CR. (1982*a*) *CTFA Ingredient Dictionary*. 3rd ed. Washington, D.C.: Cosmetic, Toiletry, and Fragrance Association.

Estrin NF, Haynes CR, Whelan JM. (1982*b*) *CTFA Compendium of Cosmetic Ingredient Composition. Specifications/Spectra*. Washington, D.C.: Cosmetic, Toiletry, and Fragrance Association.

Farsund T. (1974) Preparation of bladder mucosa cells for micro-flow fluorometry. *Virchows Arch [Cell Pathol]* 16:35–42.

Farsund T. (1978) Cell kinetics of mouse urinary bladder epithelium. VI. Changes in the proportions of cells with various nuclear DNA content after repeated doses of propylene glycol (1,2-Propanediol). *Virchows Arch [Cell Pathol]* 27:1–6.

Farsund T. (1981) Disturbances of growth and polyploidy in mouse bladder epithelium by systemic injection of toxic and carcinogenic substances. *Acta Pathol Microbiol Scand [A]* 274:304–6.

Fastner Z. (1981) Miscellaneous drugs (side effects). *Side Effects Drugs Annu* 5:436–42.

Federal Register. (January 28, 1992) Modification in voluntary filing of cosmetic product ingredient and cosmetic raw material composition statements. Final rule. 57:3128–30.

Fish KJ, Rice SA, Margary J. (1988) Contrasting effects of etomidate and propylene glycol upon enflurane metabolism and adrenal steroidogenesis in Fischer 344 rats. *Anesthesiology* 68:189–93.

Fisher AA. (1990) Use of glycerin in topical minoxidil solutions for patients allergic to propylene glycol. *Cutis* 45:81–2.

Fisher AA, Branaccio RR. (1979) Allergic contact sensitivity to propylene glycol in a lubricant jelly. *Arch Dermatol* 115:1451.

Fisher AA, Pascher F, Kanof NB. (1971) Allergic contact dermatitis due to ingredients of vehicles. *Arch Dermatol* 104:286–90.

Fligner CL, Jack R, Twiggs GA, Raisys VA. (1985) Hyperosmolality induced by propylene glycol. A complication of silver sulfadiazine therapy. *JAMA* 253:1606–9.

Floersheim GL. (1985) Protection against acute ethanol toxicity in mice by zinc aspartate, glycols, and pyritinol. *Agents Actions* 16:580–4.

Food and Drug Administration. (1984) Cosmetic product formulation data: ingredients used in each product category. Computer printout. Washington, D.C.

Food and Drug Administration. (December 2, 1991) OTC drug review ingredient status report. Food and Drug Administration, Rockville, MD.

Food and Drug Administration. (1992) Food additive safety profile. Section E. Safety of the food additive Polypropylene Glycol 1200. Submitted by FDA: FOI request dated 5/4/92.

Food and Drug Administration. (1993) Cosmetic product formulation data. FDA computer printout.

Food Chemicals Codex. (1981) 3rd ed. National Academy of Sciences, 233–4, 255–6.

Fort FL, Heyman IA, Kesterson JW. (1984) Hemolysis study of aqueous polyethylene glycol 400, propylene glycol and ethanol combinations *in vivo* and *in vitro*. *J Parenter Sci* 38:82–7.

Frosh PJ, Pekar U, Enzmann MD. (1990) Contact allergy to propylene glycol. Do we use the appropriate test concentration? *Dermatol Clin* 8:111–3.
Fujino H, Chino T, Imai T. (1965) Experimental production of labial and lingual carcinoma by local application of 4-nitroquinoline *N*-oxide. *JNCI* 35:907–18.
Gaunt IF, Carpanini FM, Grasso P, Lansdown AB. (1972) Long-term toxicity of propylene glycol in rats. *Food Cosmet Toxicol* 10:151–62.
Gebhardt CO. (1968) The teratogenic action of propylene glycol (propane-1,2-) and propanediol-1,3 in the chick embryo. *Teratology* 1:153–61.
Glasgow AM, Boeckx RL, Miller MK, et al. (1983) Hyperosmolality in small infants due to propylene glycol. *Pediatrics* 72:353–5.
Green S. (1977) Present and future uses of mutagenicity tests for assessment of the safety of food additives. *J Environ Pathol Toxicol* 1:49–54.
Guillot JP, Gonnet JF. (1985) The epicutaneous maximization test. *Curr Probl Dermatol* 14:220–47.
Guillot JP, Gonnet JF, Clement C, Caillard L, Truhaut T. (1982*a*) Evaluation of the ocular irritation potential of 56 compounds. *Food Chem Toxicol* 20:573–82.
Guillot JP, Gonnet JF, Clement C, Caillard L, Truhaut R (1982*b*) Evaluation of the cutaneous irritation potential of 56 compounds. *Food Chem Toxicol* 20:563–72.
Guillot JP, Gonnet JF, Clement C, Faccini JM. (1983) Comparative study of methods chosen by the Association Francaise de Normalisation (AFNOR) for evaluating sensitizing potential in the albino guinea pig. *Food Chem Toxicol* 21:795–805.
Halasez NA, Collins GM. (1984) Studies in cryoprotection II: propylene glycol and glycerol. *Cryobiology* 21:144–7.
Hannuksela M, Forstrom L. (1978) Reactions to peroral propylene glycol. *Contact Dermatitis* 4:41–5.
Hannuksela M, Kousa M, Pirila V. (1976) Allergy to ingredients of vehicles. *Contact Dermatitis* 2:105–10.
Hannuksela M, Pirila V, Salo OP. (1975) Skin reactions to propylene glycol. *Contact Dermatitis* 1:112–6.
Hannuksela M, Salo H. (1986) The repeated open application test (ROAT). *Contact Dermatitis* 14:221–7.
Hanzlik PJ, Mewman HW, Van Winkle W, Lehman AJ, Kennedy NK. (1939) Toxicity, fate and excretion of propylene glycol and some other glycols. *J Pharmacol Exp Ther* 67:101.
Hayashi M, Kishi M, Sofuni T, Ishidate M. (1988) Micronucleus tests in mice on 39 food additives and eight miscellaneous chemicals. *Food Chem Toxicol* 26:487–500.
Hickman JR. (1965) Acute toxicity of radiation sterilized propylene glycol. *J Pharm Pharmacol* 17:255–6.
Hickman JA, Rogers QR, Morris JG. (1990) Effect of diet on Heinz body formation in kittens. *Am J Vet Res* 51:475–8.
Hoenig V, Werner F. (1980) Is propylene glycol an inert substance? *Toxicol Lett* 5:389–92.
Huriez A, Martin P, Vanoverschelde M. (1966) L'allergie au propylene-glycol. *Bull Soc Fr Dermatol Syph* 73:263–7.
Informatics, Inc. (1973) Scientific literature review on generally recognized as safe (GRAS) food ingredients, propylene glycol and derivatives. Prepared for FDA under contract no. 72-104. NTIS no. PB-221 233.
Ingram AJ, Grasso P. (1975) Patch testing in the rabbit using a modified human patch test method. *Br J Dermatol* 109:58–9.
Instituto Ganassini Sna di Ricerche Biochimiche. (1992) Submission of unpublished data by CTFA. UV spectral analysis of propylene glycol. (1 page).*
Ishidate M, Sofuni T, Yoshikawa K, et al. (1984) Primary mutagenicity screening of food additives currently used in Japan. *Food Chem Toxicol* 22:623–36.
Jacaruso RB, Barletta MA, Carson S, Trombetta LD. (1985) Release of histamine from rat peritoneal cells in vitro as an index of irritation potential. *J Toxicol Cutan Ocul Toxicol* 4:39–48.
Kaldor G, Hsu Q, Wachsberger P, Chowrashi P. (1971) Effect of polyethylene glycols on contraction of various muscle models. *Am J Physiol* 220:639–45.
Kavlock RJ, Short RD, Chernoff N. (1987) Further evaluation of an *in vivo* teratology screen. *Teratogenesis Carcinog Mutagen* 7:7–16.
Kelner MJ, Bailey DN. (1985) Propylene glycol as a cause of lactic acidosis. *J Anal Toxicol* 9:40–2.
Kero M, Hannuksela M. (1980) Guinea pig maximization test, open epicutaneous test and chamber test in induction of delayed contact hypersensitivity. *Contact Dermatitis* 6:341–4.
Kinnunen T, Hannuksela M. (1989) Skin reactions to hexylene glycol. *Contact Dermatitis* 21:154–8.
Klecak G, Geleik H, Frey JR. (1977) Screening of fragrance materials for allergenicity in the guinea pig. I. Comparison of four testing methods. *J Soc Cosmet Chem* 28:53.

Kohn KW, Friedman CA, Ewig RA, Igbol ZM. (1974) DNA chain growth during replication of asynchronous L1210 cells. Alkaline elution of large DNA segments from cells lysed on filters. *Biochemistry* 13:4134–9.

Kulick MI, Lewis NS, Basnal, V, et al. (1980) Hyperosmolality in the burn patient: analysis of an osmolal discrepancy. *J Trauma* 20:223–8.

Lammintausta K, Kalimo K, and Fagerlund V-L. (1992) Patch test reactions in atopic patients. *Contact Dermatitis* 26:234–40.

Lashmar UT, Hadgraft J, Thomas N. (1989) Topical application of penetration enhancers to the skin of nude mice: a histopathological study. *J Pharm Pharmacol* 41:118–22.

Layton DW, Mallon BJ, Rosenblatt DH, Small MJ. (1987) Deriving allowable daily intakes for systemic toxicants lacking chronic toxicity data. *Regul Toxicol Pharmacol* 7:96–112.

Lolin Y, Francis DA, Flanagan RJ, Little P, Lascelles PT. (1988) Cerebral depression due to propylene glycol in a patient with chronic epilepsy—the value of the plasma osmolal gap in diagnosis. *Postgrad Med* 64:610–13.

Ludvigsen CW, Thurn JR, Pierpont GL, Eckfeldt JH. (1983) Kinetic enzymic assay for D(−)-lactate, with use of a centrifugal analyzer. *Clin Chem* 29:1823.

MacDonald MG, Getson PR, Glasgow AM, Miller MK, Johnson EL. (1987) Propylene glycol: increased incidence of seizures in low birth weight infants. *Pediatrics* 79:622–5.

Magnusson B, Kligman A. (1969) The identification of con-contact allergens by animal assay. The guinea pig maximization test. *J Invest Dermatol* 52:268.

Maguire HC. (1973) The bioassay of contact allergens in the guinea pig. *J Soc Cosmet Chem* 24:151.

Martin G, Finberg L. (1970) Propylene glycol: a potentially toxic vehicle in liquid dosage form. *J Pediatr* 77:877–8.

Marzulli FN, Maibach HI. (1973) Antimicrobials. Experimental contact sensitization in man. *J Soc Cosmet Chem* 24:399–421.

Maurer T, Thomann P, Weirich EG, Hess R. (1975) The optimization test in the guinea pig. A method for the predictive evaluation of the contact allergenicity of chemicals. *Agents Actions* 5:174.

Mendelow AY, Forsyth A, Florence AT, Baillie AJ. (1985) Patch testing for nickel allergy. The influence of the vehicle on the response rate to topical nickel sulfate. *Contact Dermatitis* 13:29–33.

Mochida K, Gomyoda M. (1987) Toxicity of ethylene glycol, diethylene glycol and propylene glycol to human cells in culture. *Bull Environ Contam Toxicol* 38:151–53.

Mochida K, Goto M, Saito K. (1983) Effects of diphenyl, *o*-phenylphenol, and 2-(4′-thiazolyl)-benzimidazole on growth of cultured mammalian cells. *Bull Environ Contam Toxicol* 31:428–31.

Morrissey RE, Lamb JC, Morris RW, et al. (1989) Results and evaluations of 48 continuous breeding reproduction studies conducted in mice. *Fund Appl Toxicol* 13:747–7.

Morshed KM, L'Helgoualch A, Nagpaul JP, Amma MKP, Desjeux JF. (1991) The role of propylene glycol metabolism in lactatemia in the rabbit. *Biochem Med Metab Biol* 46:145–51.

Morshed KM, Nagpaul JP, Majumder S, Amma MK. (1988) Kinetics of propylene glycol elimination and metabolism in the rat. *Biochem Med Metab Biol* 39:90–7.

Motoyoshi K, Nozawa S, Yoshimura M, Matsuda K. (1984) Safety of propylene glycol and other humectants. *Cosmet Toil* 99:83–91.

Nater JP, Baar AJ, Hoedemaeker PJ. (1977) Histological aspects of skin reactions to propylene glycol. *Contact Dermatitis* 3:181–5.

Nelson EB, Egan JM, Abernethy DR. (1987) The effect of propylene glycol on antipyrine clearance in humans. *Clin Pharmacol Ther* 41:571–3.

Nikitakis JM. (ed.) (1988) *Cosmetic Ingredient Handbook*. Washington, D.C.: Cosmetic, Toiletry, and Fragrance Association, 344, 355.

Nikko Chemicals Co., Ltd. (1991–92) *Cross Reference Index. An Index Cross-Referencing the English Names of Cosmetic Ingredients in Japan with CTFA Adopted Names. VI.* Tokyo: Nikko Chemicals Co., 20–1.

Nomura T. (1977) Similarity of the mechanism of chemical carcinogen-initiated teratogenesis and carcinogenesis in mice. *Cancer Res* 37:969–73.

National Technical Information Service. (1979) Investigation of selected potential environmental contaminants: ethylene glycol, propylene glycols and butylene glycols. Report no. PB80-109119.

Oleffe JA, Blondeel A, de Coninck A. (1979) Allergy to chlorocresol and propylene glycol in a steroid cream. *Contact Dermatitis* 5:53–4.

Patel D, Welsh D, Baker M. (1985) Comparative study of propylene glycol and caprylic/capric triglyceride vehicles for topical application. *J Soc Cosmet Chem* 36:303–11.

Pegg DE, Jacobson IA, Diaper MP, Foreman J. (1987) Perfusion of rabbit kidneys with solutions containing propane-1,2-diol. *Cryobiology* 24:420–8.

Pienta RJ. (1980) Evaluation and relevance of the Syrian hamster embryo cell system. *Appl Methods Oncol* 3:149–69.
Quinn DA, Robinson D, Hales CA. (1990) Intravenous injection of propylene glycol causes pulmonary hypertension in sheep. *J Appl Physiol* 68:1415–20.
Ramstad T, Nestrick TJ, Stehl RH. (1978) Determination of polypropylene glycol extracted from polymers into food-simulating solvents. *Anal Chem* 50:1325–7.
Romaguera C, Garcia-Perez A, Moran M, et al. (1981) Propylene glycol in standard patch tests. *Contact Dermatitis* 7:346.
Rothschild L, ed. (1988) *The Food Chemical News Guide to the Current Status of Food Additives and Color Additives*. Washington, D.C.: Food Chemical News, 372.2.
Ruddick JA. (1972) Toxicology, metabolism, and biochemistry of 1,2-propanediol. *Toxicol Appl Pharmacol* 21:102–11.
Saini M, Meenakshi, Morshed KM, Amma MKP. (1987) Propane-1,2-diol induced changes in plasma proteins and enzymes on acute oral ingestions in female rats. *Res Bull Panjab Univ Sci* 38:79–86.
Sasaki M, Sugimura K, Yoshida MA, Abe S. (1980) Cytogenetic effects of 60 chemicals on cultured human and Chinese hamster cells. *Kromosomo (Tokyo)* 20:574–84.
Sax NI. (1979) *Dangerous Properties of Industrial Materials*. New York: Van Nostrand Reinhold, 943.
Shaffer CB, Carpenter CP, Critchfield FH, Nair JH, Franke FR. (1951) A toxicological study of some polypropylene glycols. *AMA Arch Ind Hyg Occup Med* 3:448–53.
Shideman FE, Procita L. (1951) Some pharmacological actions of polypropylene glycols of average molecular weight 400, 750, 1200, and 2000. *J Pharmacol* 103:293–305.
Singh PP, Junnarkar AY, Seshagirirao C, et al. (1982) A pharmacological study of propane-1,2-diol. *Arzneimittelforsch* 32:1443–6.
Speth PA, Vree TB, Neilen NF, et al. (1987) Propylene glycol pharmacokinetics and effects after intravenous infusion in humans. *Ther Drug Monit* 9:255–8.
Staples R, Misher A, Wardell JJ. (1967) Gastrointestinal irritant effect of glycerol as compared with sorbitol and propylene glycol in rats and dogs. *J Pharm Sci* 56:398–400.
Stenback F, Shubik P. (1974) Lack of toxicity and carcinogenicity of some commonly used cutaneous agents. *Toxicol Appl Pharmacol* 30:7–13.
Sweet DV. (1987) *Registry of Toxic Effects of Chemical Substances*. Washington, D.C.: U.S. Government Printing Office.
Swenberg JA, Petzold GL, Harbach PR. (1976) In vitro DNA damage/alkaline elution assay for predicting carcinogenic potential. *Biochem Biophys Res Commun* 72:732–8.
Thomas PA, Bhat KS, Kotian KM. (1980) Antibacterial properties of dilute formocresol and eugenol and propylene glycol. *Oral Surg* 49:166–70.
Trancik RJ, Maibach HI. (1982) Propylene glycol: irritation or sensitization? *Contact Dermatitis* 8:185–9.
Van Winkle W, Newman HW. (1941) Further results of continued administration of propylene glycol. *Food Res* 6:509–16.
Wahlberg JE. (1984) Erythema-inducing effects of solvents following epicutaneous administration to man—studied by laser Doppler flowmetry. *Scand J Work Environ Health* 10:159–62.
Wahlberg JE, Nilsson G. (1984) Skin irritancy from propylene glycol. *Acta Derm Venereol (Stockh)* 64:286–90.
Wakabayashi T, Adachi K, Popinigis J. (1991) Effects of alkyl alcohols and related chemicals on rat liver structure and function. 1. Induction of two distinct types of megamitochondria. *Acta Pathol Jpn* 41:405–13.
Wallenius K, Lekholm U. (1973*a*) Oral cancer in rats induced by the water-soluble carcinogen 4-nitrochinoline *N*-oxide. *Odont Revy* 24:39–47.
Wallenius K, Lekholm U. (1973*b*) Influence of saliva on epidermal cancer in rat induced by water or fat-soluble carcinogens. *Odont Revy* 24:115–26.
Warshaw TG, Herrmann F. (1952) Studies of skin reactions to propylene glycol. *J Invest Dermatol* 19:423–30.
Weast RC. (1982) *CRC Handbook of Chemistry and Physics*. Boca Raton: CRC Press, C-480.
Weil CS, Woodside MD, Smyth HF, Carpenter CP. (1971) Results of feeding propylene glycol in the diet to dogs for two years. *Food Cosmet Toxicol* 9:479–82.
Wilkinson DS, Fregert S, Magnusson B, Bandmann HJ, Calnan CD. (1970) Terminology of contact dermatitis. From the International Contact Dermatitis Research Group. *Acta Derm Venereol (Stockh)* 50:287–92.
Willis CM, Stephens JM, Wilkinson JD. (1988) Experimentally-induced irritant contact dermatitis. Determination of optimum irritant concentrations. *Contact Dermatitis* 18:20–4.

Willis CM, Stephens CJ, Wilkinson JD. (1989) Epidermal damage induced by irritants in man: a light and electron microscopic study. *J Invest Dermatol* 93:695–9.

Willis CM, Stephens CJM, Wilkinson JD. (1990) Differential effects of structurally unrelated chemical irritants on the density and morphology of epidermal CD 1+ cells. *J Invest Dermatol* 95:711–6.

World Health Organization. (1974) *Seventeenth Report of the Joint Food and Agriculture Organization/WHO Expert Committee on Food Additives: Toxicological Assessment of Certain Food Additives with a Review of General Principles and Specifications. Report No. 539.*

Yu DK, Sawchuk RJ. (1987) Pharmacokinetics of propylene glycol in the rabbit. *J Pharmacokinet Biopharm* 15:453–72.

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